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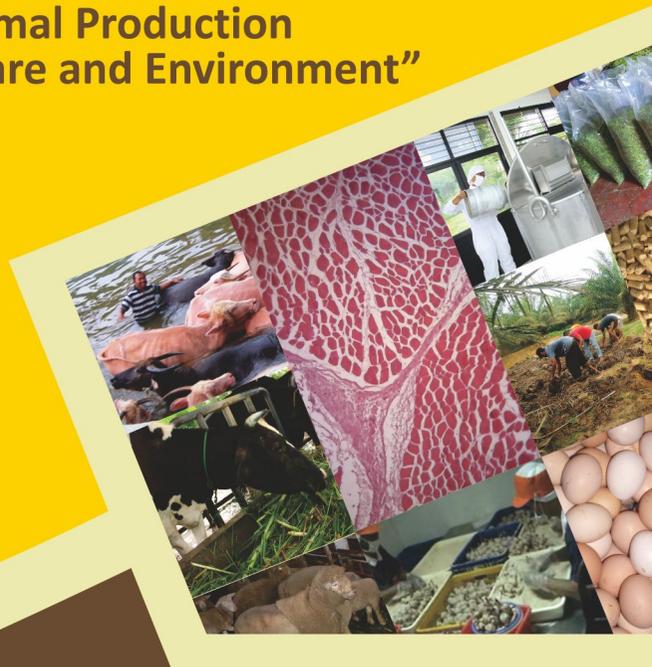
# **PROCEEDING**



**THE THIRD INTERNATIONAL SEMINAR ON ANIMAL INDUSTRY**

**“Sustainable Animal Production  
for Better Human Welfare and Environment”**

September, 17-18 2015  
IPB International Convention Center  
Bogor-Indonesia



**Organized by:**



**Sponsored by:**



**FACULTY OF ANIMAL SCIENCE  
BOGOR AGRICULTURAL UNIVERSITY  
2015**

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Proceeding of the 3<sup>rd</sup> International Seminar on Animal Industry,  
Bogor, 17-18 September 2015

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# FOREWORD FROM CHAIRPERSON OF ORGANIZING COMMITTEE

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Distinguished,

Rector of Bogor Agricultural University, Prof. Dr.Ir. Herry Suhardiyanto, M.Sc.

Director General of Livestock Services and Animal Health, Ministry of Agriculture, Republic of Indonesia, Prof. Dr. Ir. Muladno, MSA.

Dean of Animal Science Faculty, Bogor Agricultural University, Prof Dr Luki Abdullah M.Agr.Sc.

All participants of the International Seminar on Animal Industry 2015

Good morning ladies and gentlemen,

It is my great pleasure to welcome you all, distinguished guests, speakers and participants, to the Third International Seminar on Animal Industry (ISAI 3rd, 2015) held at the IPB International Convention Center, Bogor Indonesia. This seminar with the theme **“Sustainable Animal Production for Better Human Welfare and Environment”** is organized by Faculty of Animal Science, Bogor Agricultural University in collaboration with Association of Indonesia Animal Scientists.

Following the recommendations from Isai 1 and Isai 2, which were held in Indonesia in 2009 and 2012, the strategic issues of Isai 3rd is emphasized on animal production systems and technology and the use of natural resources in relation with environmental aspects, toward a sustainable animal production. There will be 97 papers presented during the two days seminar; 9 by invited speakers, 69 for oral and 28 for posters presentations. The speakers came from different countries including Australia, Egypt, France, Korea, German, Netherland, Indonesia, Malaysia, Nigeria, Pakistan, Thailand, USA.

This is a great opportunity for scientists, researchers, private sectors and policy makers to discuss, share information and experiences on interesting topics in animal production in a broad sense, including good farming practices, recent technologies and save animal products. I believe, there is an open window for initiating and strengthening collaboration among scientists and institutions during and after the seminar.

On behalf of the Organizing Committee, I would like to express my sincere appreciation and thanks to IPB, and some units within, including Institute of Research and Community Empowerment, Faculty of Animal Science, Department of Animal Production and Technology, Department of Nutrition and Feed Technology, Diploma Program, Management and Business Program for all advice and funding support.

The success of this seminar could only be achieved with all the valuable supports and sponsorship from some recognized institutions in this country. In this regards, I would like to address my grateful thanks to Directorate General of Livestock Services and Animal Health-Indonesia Ministry of Agriculture for funding support, and Infovet and Trobos, Green TV as promotion agency. To: Sierad Produce, Kaltim Prima Coal, BRIngin Life, Adaro Indonesia, Trouw Nutrition Indonesia, Nutricell Pasific, Sweni Transfer Indonesia, Charoen Phokphand, Wide & Pin, Pupuk Kujang, and ANTAM thank you so much with big appreciation, for having being part of this important event and such enormous contributions.

My recognition and gratitude are also forwarded to the Steering Committee for advice and assistanship, to international and national reviewers and the Scientific Committee for hard working and such great contribution. Last but not least, to all my dear colleagues of the Organizing Committee members, who have been working smartly and full of dedication and passion, to make this seminar a great successful event.

To all participants, hopefully, the two days seminar may bring fresh ideas, and enhancing collaborations for future success toward sustainable animal production. Big apologies for any inconveniences during the seminar, wish you all having good times, and fruitful discussions.

During your short stay, please enjoy the surrounding of Bogor city, the Museum of Presidential Palace and Historical Botanical Garden of Bogor.

Bogor, September 17th, 2015

The Isai 3rd 2015,

Chairperson of Oraganizing Committee

Asnath M. Fuah

## REMARKS FROM DEAN OF ANIMAL SCIENCE FACULTY

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Prof. Dr. Muladno, MSA

Director General of Livestock and Animal Health-Ministry of Agriculture Republic of Indonesia,

Prof. Dr. Ir. Herry Suhardiyanto, M.Sc.

Rector of IPB

Dr. Ir. Asnath Maria Fuah

Chairperson, The 3<sup>rd</sup> International Seminar on Animal Industry

Our Colleagues from Indonesian universities and research institutes,  
Distinguished foreign participants and speakers,

Representative of livestock services officers of local government from all over Indonesia,

Distinguished guests, ladies and gentlemen.

Assalamu'alaikum warahmatullaahi wabarakatuh,

I am pleased to welcome you all to Bogor city for attending "The 3<sup>rd</sup> International Seminar on Animal Industry 2015" held at Faculty of Animal Science, Bogor Agricultural University (IPB). As the Dean of Faculty, I am also really honored to host this conference.

First, let me introduce briefly about Bogor city. Bogor is one of the major scientific and educational centers in Indonesia. A significant part of academic and research base was laid in the period of Dutch colonization. In particular, since the beginning of the 19<sup>th</sup> century there were established laboratories and professional schools focused primarily on improving the efficiency of the colonial agriculture. Similar to the prevailing profile of research and academic activity was retained in Bogor after gaining independence. As in the second half of 20<sup>th</sup> century, and in the 2000s strongest areas were Agricultural sciences, Biology, Animal and Veterinary Sciences. The main educational and scientific center with the utmost national importance is the Bogor Agricultural University (IPB). It is therefore the city regularly hosted various international events, such as international seminars and conferences.

I would like to express my gratitude to IPB for supporting us to hold this conference, and also to the organizing committee of the present conference for their hard work and persistence. I convey my sincere gratitude to all the parties which is supporting this event, such as Directorate General of Livestock and Animal Health-Ministry of Agriculture Republic of Indonesia, Infovet Trobos, Agrina, Green TV as promotion agency and Sierad Produce, Kaltim Prima Coal, BRIngin Life, Adaro Indonesia, Trouw Nutrition Indonesia, Nutricell Pasific, Sweni Transfer Indonesia, Charoen Phokphand, Wide & Pin, Pupuk Kujang, and ANTAM thank you so much with big appreciation, for having being part of this important event and such enormous contributions. I am very pleased to see here the delegates from various foreign countries as well as representatives from many domestic institutions.

I hope you find this conference and the city, both interesting and stimulating and that you enjoy meeting up with your professional colleagues as well as having pleasure time during your stay in Bogor.

Thank you very much and

Wassalamu'alaikum warahmatullaahi wabarakaatuhu.

Bogor, 17 September 2015

Prof. Dr. Ir. Luki Abdullah, MSc.Agr

DEAN



# SEMINAR PROGRAM

## Conference Program Thursday, September 17, 2015

Time Slot	Venue : ICC Ballroom	
	Event	Speaker
08.00-09.00	Registration	Committee
09.00-09.05	Opening Ceremony	Master of Ceremony
09.05-09.15	Report from Organizing Committee	Dr. Ir. Asnath M.Fuah, MS
09.15-09.25	Welcome Address from Dean Faculty of Animal Science	Prof. Dr. Ir. Luki Abdullah, M.Sc.Agr.
09.25-09.35	Welcome Address from Rector of Bogor Agricultural University	Prof. Dr. Ir. Herry Suhardiyanto, M.Sc
09.35-10.00	Opening and Keynote Speech by Ministry of Agriculture / Directorate General of Livestock and Health Services	Prof. Dr. Ir. Muladno, MSA
10.00-10.05	Appreciation for Keynote Speakers from Dean Faculty of Animal Science	Prof. Dr. Ir. Luki Abdullah, M.Sc.Agr.
10.05-10.20	Sponsorship Appreciation from Chairman of Organizing Committee	Dr. Ir. Asnath M.Fuah, MS.
10.20-10.25	Photo session	Photographer
10.25-10.40	Coffee break	
	Plenary Session 1 <i>Moderator: Prof. Dr. Ir. Komang G. Wiryawan</i>	
10.40-11.00	Invited speaker 1	<b>Prof. Dr. Ir. Bas. Kemp</b> Preserving Health, Welfare and Productivity in a Challenging Environment
11.00-11.20	Invited speaker 2	<b>Dr. Jean Pierre Bidanel</b> Genomic Selection for More Sustainable Livestock Production
11.20-11.40	Invited speaker 3	<b>Ir. Yunus Triyonggo, MM</b> Building Human Resources Competency Model in Poultry Industry
11.40-12.00	Discussion	
12.00-12.05	Invited Speaker Appreciation from Scientific Committee	Prof. Dr. Ir. Dewi Apri Astuti, MS.
12.05-12.15	Sponsorship Appreciation from Vice Dean Faculty of Animal Science	Dr. Ir. Moh. Yamin, M.Agr.Sc.
12.15-12.25	Student Plenary	
12.25-13.20	Lunch	
13.20-13.50	Poster session	

<b>Time</b>	<b>Room A (Theme D)</b>	<b>Room B (Theme B and C)</b>
<b>Session 1</b>	<b>Moderator : Dr. Rajesh Jha</b>	<b>Moderator : Ummi Noorhakimah</b>
14.00-14.10	<b>Thongsuk Jetana</b> Rain Tree Pod in Livestock Feeds: Opportunity, Challenges and Possibility	<b>Yeni Widiawati</b> Fermentation Kinetics Of Palm Oil Plantation By-Product Based Diet
14.10-14.20	<b>Supriyati Kompiang</b> Effect of Different Protein and Energy Levels in Concentrate Diets on Performances of Anglo-Nubian Goat During Pregnancy and Lactation Periods	<b>Ainissya Fitri</b> Utilization Of Haylage Of Local Agro- Industry By product Pretreated With Afex Method
14.20-14.30	<b>Rusdi</b> Evaluation of Eleutherine ( <i>Eleutherine americana</i> ) as Feed Additive for Poultry	<b>H. A. Sukria</b> Physical Quality And Storage Time Pellet Indigofera Spleaves
14.30-14.40	Discussion	Discussion
<b>Session 2</b>	<b>Moderator :Thongsuk Jetana</b>	<b>Moderator : Imana Martaguri</b>
14.45-14.55	<b>Utsav Prakash Tiwari</b> Nutrient Profile And In Vitro Digestibility Of Fresh And Ensiled Cassava In Swine	<b>Moh Ali Hamdan</b> Potential Of Dwarf Elephant Grass ( <i>Pennisetum Purpureum</i> Schum. Cv. Mott) In Dry Land Areas Of Bojonegoro As Forage- Based Feed Sustainability
14.55-15.05	<b>Alif Putri</b> Effect of Combination Silkworm Pupae Meal and Garlic Meal on Blood Profiles, Visceral Organs and Carcass Broiler	<b>Rido Pande Pardede</b> Development Of Indigofera Zoolingeriana And Pueraria Javanica On Dry Land Integrated With Teak Forest In Bojonegoro
15.05-15.15	<b>Burhanudin Sundu</b> The effect of NaOH Concentrations and Polysaccharides Extract of Palm Kernel Meal on Performance of 4 Weeks Old-Broiler Chickens	<b>Malcky Telleng</b> Growth and Productivity of Different Sorghum Varieties Cultivated with Indigofera in Intercropping System
15.15-15.25	Discussion	Discussion
15.25-15.40	Coffee break	
<b>Session 3</b>	<b>Moderator : Anis Mukhtiani</b>	<b>Moderator : Lisa T. Praharani</b>
15.40-15.50	<b>Muhamad Nasir Rofiq</b> Combination Effect of Nutritech Feed Additive Containing Saponin, Tanin and Eugenol Essential oils on In Vivo Rumen Methane Production in Dairy Cattle Using Open Circuit Respiration Chamber Technique	<b>Imana Martaguri</b> Carbon Storage Capacity of Forage Native Grasses Growing in Palm Plantation at Transformation Forest Ecosystem in Jambi
15.50-16.00	<b>Dwi Yulistiani</b> Nitrogen Utilization and Ruminant Fermentation of Five Breed of Sheep Fed Concentrate Containing Different Levels of Rumen Undegradable Protein	<b>I Gusti Ngurah Jelantik</b> Herbage Production and Nutritive Value of Some Forage Legumes as Calf Feed Supplement
16.00-16.10	<b>Sutresniwati</b> A Willingness to Pay Evaluation for Silage Implementation for Small Dairy Farmers	<b>Riesi Sriagtula</b> Evaluation of Growth and Production of Sorghum Lines (Sorghum Brown Midrib) at Different of Harvest Time as Feed
16.10-16.20	Discussion	Discussion
<b>Session 4</b>	<b>Moderator : Rusdi</b>	<b>Moderator: Veronica</b>
16.25-16.35	<b>Anita S. Tjakradidjaja</b> Fermentability and Digestibility of Rice Straw - Concentrate Base Ration Added with Probiotic	<b>Nur Rochmah Kumalasari</b> Modelling of Forage Availability Response to Landuse Exchange in Bogor

Time	Room A (Theme D)	Room B (Theme B and C)
16.35-16.45	<b>Gusti A. Gultom</b> Effects of Solid or Liquid Probiotic Supplementation on Rumen Microbial Population and Enzyme Activity	<b>Khalil</b> The Diversity and Quality of Forages Used for Feeding of Goat in Payakumbuh of West Sumatra
16.45-16.55	<b>Eissa M. M</b> Effect Of Ammoniated Straw On Methane Production In An In Vitro System And On Growth Performance	<b>P.D.M.H. Karti</b> The Addition of Arbuscular mycorrhizal Fungi in Enhancing Productivity and Drought Tolerance Mechanisms of <i>Indigofera zollingeriana</i>
16.55-17.05	Discussion	Discussion

Time	Ballroom (Theme A)
<b>Session 1</b>	<i>Moderator : Iis Arifiantini</i>
14.00-14.10	<b>Fuah A.M</b> Beef Cattle Production, Constraints and Opportunities for Small Farmers in South Central Timor Regency West Timor
14.10-14.20	<b>S.N. Sirajuddin</b> The Application of Tesang Sharing System at Cattle Farms in Indonesia
14.20-14.30	<b>Niken Ulupi</b> Production Performance of Laying Hen in Cage System with Different Housing Temperature
14.30-14.40	<b>Lucia Cyrilla</b> Evaluation of Good Dairy Farming Practice Implementation in Dairy Goat Farm
14.40-14.50	Discussion
<b>Session 2</b>	<i>Moderator : Prof. Cece Sumantri</i>
14.55-15.05	<b>Lindawati Doloksaribu</b> Constraints to, Challenges of, and Opportunities for Rearing Goats in Bali Province. A case study: Rearing Kids in Karangasem Regency
15.05-15.15	<b>Hearty Salatnaya</b> Trigona Spppropolis, Pollen, And Honey Production In Two Different Agroecosystem
15.15-15.25	<b>Prabowo, S</b> Distribution of Thermal Body Surface Ettawah Grade in Different Tropic Microclimates
15.25-15.35	<b>Bram Brahmantiyo</b> Hycole and Hyla Rabbits Performance were Raised in Indonesia
15.35-15.45	Discussion
15.45-16.00	<b>Coffee break</b>

### Welcoming dinner. Venue ICC Ballroom

Time Slot	Event
18.20-19.00	Registration and Dinner (Instrument from Gentra)
19.00-19.05	Opening by Master of Ceremony
19.05-19.15	Speech from Chairman of Committee
19.15-19.25	Speech from Dean of Animal Science Faculty
19.25-20.00	Gentra Kaheman
20.00-20.20	Prof. Singer
20.20-21.20	Spontaneity from Country Representative
21.20	Closing

Friday, September 18, 2015

Time	Venue : IICC Ballroom	
	Event	Speaker
8.00-8.30	Registration	Committee
8.30-8.35	Opening Ceremony	Master of Ceremony
	Plenary Session 2 <i>Moderator: Dr. Jean Pierre Bidanel</i>	
8.35-8.55	Invited speaker 1	<b>Prof. Wayne Pitchford</b> Outcomes of Selection for Residual Feed Intake in Australian Beef Cattle
8.55-9.15	Invited speaker 2	<b>Prof. Myunggi Baik</b> Molecular Mechanisms Regulating Beef Quality in Korean Cattle
9.15-9.35	Invited speaker 3	<b>Prof. I Wayan Teguh W.</b> Vaccination and Subclinical Manifestation of Avian Influenza in Indonesia
9.35-9.50	Discussion	
9.50-10.00	Appreciation to Invited Speaker	Prof. Luki Abdullah
10.00-10.10	Coffee Break	
	Plenary Session 3 <i>Moderator: Prof. Wayne Pitchford</i>	
10.10-10.30	Invited speaker 1	<b>Dr. Kai J. Kuehlmann</b> The Role of Feed Additive in Animal Industry under Tropical Condition
10.30-10.50	Invited speaker 2	<b>Dr. Anjas Asmara Samsudin</b> Recent Advances in Gut Microbiology Research in Relation to Animal Nutrition
10.50-11.10	Invited speaker 3	<b>Prof. Bustanul Arifin</b> Social Economic and Policy in Animal Industry
11.10-11.25	Discussion	
11.25-11.30	Appreciation for Invited Speaker	Prof. Dr. Ir. Sumiati, M.Sc.
11.30-13.20	Lunch and Prayer	
13.20-13.50	Poster session	

Time	Room A (Theme D and G)	Room B (Theme F and J)
<b>Session 5</b>	<i>Moderator: Sutresniwati</i>	<i>Moderator : Dr. Irma Isnafia Arif</i>
13.50-14.00	<b>Sumiati</b> Effect of drinking gambir extract ( <i>Uncaria gambir Roxb</i> ) as Antioxidant on Performance of 40-43 Weeks Old of Laying Hens	<b>Rudi Afnan</b> Weight Loss And Mortality Of Broiler During Transportation From Different Distances To Slaughterhouse
14.00-14.10	<b>Muktiani, A</b> Live Weight Gain of Beef Cattle Fed on Complete Feed Silage of Water Hyacinth Supplemented with Mineral Zinc-Proteinate	<b>Hajrawati</b> Meat Quality Of Marica Goat ( <i>Capra Hircus</i> ) Meat Fed Different Protein Level
14.10-14.20	<b>Putri O. N</b> The Effect of Adding Fermented Waste Cabbage in Calf Starter Pellets on Total Lactic Acid Bacteria And <i>Escherichia coli</i>	<b>Suharyanto</b> Skim Milk Powder Substitution With Soymilk Powder Could Improve Physical Properties Of Beef Surimi-Based Sausage
14.20-14.30	Discussion	Discussion
<b>Session 6</b>	<i>Moderator : Prof. Khalil.</i>	<i>Moderator : Salina AB</i>
14.35-14.45	<b>Ninasari Ra</b> Substitution of Fish Meal by Cricket or Indigofera Shoot Leaf Meal on Japanese Quail ( <i>Coturnix japonica</i> ) Performance	<b>Lilis Suryaningsih</b> Effects Of Local Flour Types On Physical Properties And Acceptability Of Beef Sausage

Time	Room A (Theme D and G)	Room B (Theme F and J)
14.45-14.55	<b>Tresia G.E</b> Benefit of Kemuning Leaves Meal ( <i>Murraya paniculata</i> [L.] Jack) Addition in Ration Containing Date Fruit Waste to Suppress Gastrointestinal Parasites Infestation of PE Goat	<b>Soenarno Ms</b> Characteristic Of Lactic Acid Bacteria Isolated From Dangke From Sinjai, South Sulawesi
14.55-15.10	<b>Sri Suharti</b> Rumen Microbe, Protein Microbial Synthesis, Celullase Activity and Nutrient Digestibility of Bali Cattle Rumen with the Addition of Calcium Soap-Soybean Oil In vitro	<b>M. Aman Yaman</b> Increase on Commercial Weight, Carcass Quality and Economic Benefit of Selected Local Meat Chicken Fed on Fermented Diet Contained Digestive Enzymes and Probiotics
15.10-15.15	Discussion	Discussion
15.15-15.30	Coffee break	
<b>Session 7</b>	<b>Moderator : Dr. Lindawati Doloksaribu</b>	<b>Moderator : Dr. Asnath Maria Fuah</b>
15.30-15.40	<b>G. F. Bira</b> Incremental Level Of Chromolaena Odorata In Complete Diet Does Not Impair Intake, Rumen Fermentation And Microbial Protein Synthesis Efficiency In Cattle	<b>Salina A.B</b> An Analysis Of Cattle Traders Practices On Animal Traceability In Malaysia
15.40-15.50	<b>Arini NMJ</b> Subtitution Of Fish Meal By Cricket Or Indigoferasp Shoot Leaf Meal To Evaluate Protein Balance Of Japanese Quail (Coturnix Japonica)	<b>Hotnida C H Siregar</b> Effect Of Moisture Reduction Method, Storage Period And Temperature On Honey Quality
15.50-16.00	<b>Mokhamad Faesal R. Hakim</b> Feeding Ecology of Sumatran Orangutan ( <i>Pongo abelii</i> , Lesson 1827) in West Batang Toru Forest Block, North Sumatra	<b>Iman Rahayu</b> Biodiversity Based On Fatty Acid And Amino Acid Profile Of Indonesian Local Chickens
16.00-16.10	Discussion	Discussion
<b>Session 8</b>	<b>Moderator : Mokhamad Faesal Rakhman Khakim</b>	<b>Moderator : Dr. Burhanudin Sundu</b>
16.15-16.25	<b>D. Latipudin</b> Level Of Malondialdehyde (Mda), Uric Acid And Lymphocyte: Neutrphyl Ratio Of Laying Hen In The Different Temperature Humidity Index (Thi)	<b>I M. A. Sudarma</b> Weight Loss Of Inter-Island Transported Cattle From Kupang Is Reduced By Feeding High Protein-Mineral Mix Block During Quarantine And Sea Transportation
16.25-16.35	<b>Windi Al Zahra</b> The Using Of Thermograph As Non-Invasive Method To Observe Subclinical Mastitis In Tropical Dairy Cattle	<b>Ummi Noorhakimah Abdullah</b> Cattle Importation And The Trend Of Fmd Occurrence In Peninsular Malaysia From 2000-2010
16.35-16.45	<b>A. Sudarman</b> Physiological Responses And Blood Profiles Of Sheep Fed Cassava Leaves Silage ( <i>Manihot Esculenta</i> Sp.) Reared Traditionally In Petir Village	<b>Moh Yamin</b> Harmony Between Livestock Behaviors: Birth Time and Sites Selection Behaviors in Sheep and Goats
16.45-17.00	Discussion	<b>Erika B Laconi</b> Strategy of Beef Cattle Development Based on Agricultural Product in Kuningan District, West Java
17.00-17.10		Discussion

<b>Time</b>	<b>Ballroom (Theme E and J)</b>
<b>Session 5</b>	<b>Moderator : Anneke Anggraeni</b>
13.50-14.00	<b>Surya Nur Rahmatullah</b> Phenotypic Variation In Male Local Chicken At Tapin Regency Using Significant Analysis
14.00-14.10	<b>Parsaoran Silalahi</b> Effects Of Selection On The Efficiency And Variability Of Sow Reproduction And Maternal Abilities
14.10-14.20	<b>Oktora Dwi Putranti</b> Effect Of Caffeine On Morfology Of Epididymis Spermatozoa Of Bali Bull
14.20-14.30	Discussion
<b>Season 6</b>	<b>Moderator : Ir Anita S.T. MRur.Sc</b>
14.35-14.45	<b>Lisa Praharani</b> Comparisson of Anglo Nubian X Etawah Grade Goats And Saanen X Etawah Grade Goats For Some Reproductive Traits
14.45-15.00	<b>Maria Haryulin Astuti</b> Service Per Conception In Beef Cattle With Artificial Insemination In Kapuas Basarang District of Central Kalimantan
15.00-15.10	<b>Anneke Anggraeni</b> Association Of Growth Hormone (Gh Mspi) And Growth Hormone Releasing Hormone (Ghrh  Haeiii) Genes With Milk Components Of Hf Cows Under Small Farmers In Lembang, West Java
15.10-15.20	Discussion
15.20-15.30	Coffee break
<b>Season 7</b>	<b>Moderator : Dr. Epi Taufik</b>
15.30-15.40	<b>R.Iis Arifiantini</b> Hypoosmotic Test In Rabbit Spermatozoa
15.40-15.50	<b>Nalley Wmm</b> Effect Of Freezing On Bovine Sperm Morphology
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16.25-16.35	<b>Aslina Asnawi</b> Financing Preferences For Cattle Farmers In Bone Regency South Sulawesi
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# **INVITED SPEAKER**

**Proceeding of the 3<sup>rd</sup> International Seminar on Animal Industry,  
Bogor, 17-18 September 2015  
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# Preserving Health, Welfare and Productivity in a Challenging Environment

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## Abstract

*Health, welfare and productivity are shaped that the animal's capacity to adapt. A 'load' of simultaneous, persistent or severe challenges exceeding the adaptive capacity of farm animals may lead to reduced resilience and culminate in behavioural and physiological disturbances and disease susceptibility. So there should be a proper balance between the challenges an animal is exposed to and their adaptive capacity. The adaptive capacity of animals is influenced by their early-life experience. Evidence is accumulating that perinatal experiences may have long-term effects on the ability to cope with environmental challenges later in life. High incubation temperatures in broilers, for instance, were demonstrated to hamper heart development with lasting effects on the susceptibility to develop ascites in later life. In early postnatal life of chickens and pigs, a timely development of nutritional independence seems important. Poultry systems that provide chicks with feed and water immediately after hatch have beneficial and long-term effects on growth and immune competence. Also in piglets, early experience with the ingestion of feed, already early in lactation, may have important developmental effects. To stimulate early feed intake in piglets, enrichment of the environment with foraging substrates and facilitating the transfer of information from the sow about what and where to eat are important. Also reduction of challenges in the current environment can balance adaptive capacity with environmental challenges. Shortening of the dry period in dairy cattle has been shown to reduce the negative energy balance in early lactation thereby improving metabolic health.*

*Keywords: adaptation, early life experiences*

## Resilience and Adaptive Capacity

Animals are striving for homeostasis to optimize growth and to reproduce. Animals are exposed to a variety of environmental factors like variation in ambient temperature, pathogens, and resource (nutrient) availability, that potentially threaten homeostasis and therefore optimal functioning. The animal is however equipped with a set of physiological systems that cope with these environmental perturbations. E.g. the animal has a thermoregulatory systems to react to heat and cold exposure and an immune system to battle invading pathogens. With these physiological systems the animal is at least partly resilient to challenges in the environment it is exposed to. Resilience can be described as the ability of an animal to react (adapt) to changing environmental conditions with minimum loss of function. Resilience is bordered by the adaptive capacity of the physiological systems of an animal to combat environmental perturbations. If environmental perturbation or challenges are outside the adaptive capacity of an animal, resilience in terms of health, welfare and productivity is at stake. A 'load' of simultaneous, persistent or severe challenges exceeding the adaptive capacity of farm animals will lead to reduced resilience and culminate in behavioural and physiological disturbances and disease susceptibility. So there should be a proper balance between the challenges an animal is exposed to and their adaptive capacity. Health, welfare and productivity are therefore shaped that the animal's capacity to adapt.

## Early Life Environmental Influences on Adaptive Capacity Later in Life (Prenatal Effects)

The adaptive capacity of animals is influenced by their early-life experience. Evidence is accumulating that perinatal experiences may have long-term effects on the ability to cope with environmental challenges later in life. Already before birth or hatch (in chickens) environmental effects may influence the offspring through effects of the environment on the mother or parents. An example of transgenerational effects can be found in laying hen. De Haas *et al.* (2014) studied effects of parent flock stress and feather pecking levels on offspring feather pecking and stress levels. In particular lines they found convincing evidence

that parent stock with high feather damage, high basal corticosterone levels and serotonin levels produced offspring that had higher levels of severe feather pecking at young age. This implies that a typical sign of non-adaptation like feather pecking finds, at least partly, its origin in stress and feather pecking levels in the mother lines. Exact mechanisms by which this information of parents is transferred to the offspring is still under study.

Another example of prenatal effects on later resilience was found in broilers where high incubation temperatures were found to increase the mortality due ascites later in life (Molenaar *et al.* 2010). Ascites is a typical metabolic syndrome found in fast growing broilers due to limitation of the cardiovascular system to keep up with the high metabolic rate of these birds. Molenaar *et al.* (2010) showed that ascites related mortality increased from 2.8 to 6.6% when egg shell temperatures were raised from 37.8 to 38.9°C during the last stage of incubation. These higher incubation temperatures are often found in practice especially in poor ventilated hot spots in the incubator and in multi stage incubator systems. More detailed studies show that high incubation temperatures in broilers hamper heart development during incubation which may reduce their cardiovascular capacity later in life. The poor heart development at high incubation temperatures is at least partly explained by the lower mitotic rate in the heart muscle at higher incubation temperatures (Romanoff 1960) and partly by an increase embryonic need for glucose at high incubation temperatures. This limits development of glycogen stores needed for hatching and results in the use of protein from the heart muscle as a glycogenic energy source to hatch (Molenaar *et al.* 2013).

Prenatal effects on later health, welfare and performance are also described other farm animals. An example is flavour learning in pigs. Weaning of piglets is a typical critical transition period in pigs where signs of maladaptation are evident by high stress levels, low nutrient intakes, poor growth rate or even weight loss, impaired intestinal functioning and a high use of antibiotics. Moreover, stress during weaning may induce maladaptive behaviours like tail biting which once learned may result in tail biting outbreaks later in life. With perinatal flavour learning, flavours are fed to sows during pregnancy and lactation which can pass to the offspring via amniotic fluid and in milk. Research has shown that perinatally exposed piglets through the sows feed have lower cortisol responses around weaning and a better growth and feed intake, lower diarrhoea incidence and less maladaptive behaviours after weaning, if the flavour was present in the post weaning environment (Oostindjer *et al.* 2009, 2010a, 2011a). The effect was most pronounced with exposure to flavour during pregnancy.

Collectively these results show that the prenatal environment can have pronounced effects on later life resilience of farm animals.

## **Early Life Environmental Influences on Adaptive Capacity Later in Life (Postnatal Effects)**

Also environmental conditions in the early postnatal environment can affect later life resilience. For example, in early postnatal life of chickens and pigs, a timely development of nutritional independence seems important.

In industry most chickens hatch in incubator and when most of the chickens have hatched they are transported to the farm where feed and water is provided for the first time. Since actual hatch moment can vary between about 460 and 500 h and transport condition may vary considerable in duration, there is substantial variation in time when feed and water is provided after hatch. However, poultry systems that provide chicks with feed and water immediately after hatch have beneficial and long-term effects on growth in broilers (Van der Ven *et al.* 2011). In layers it was shown that feeding immediately after hatch has long term effects on intestinal microbiota composition and increases systemic humoral T-cell dependent immune responses (Walstra 2011, Kingma, unpublished results). It appears that early feed and water provision in chickens affect performance and immune reactivity later in life.

Also in piglets, early experience with the ingestion of feed, already early in lactation, may be have important developmental effects. It has been shown in many studies (e.g. Kuller *et al.* 2004) that solid feed intake of piglets before weaning is positively related to feed intake after lactation. So stimulation of feed intake before weaning may contribute to an easier transition through weaning. In general, even if creep feed is available from early lactation onwards, actual feed intake of piglets is highly variable between and within litters. So the question is how piglets can learn what, where and how to eat already during lactation. To stimulate early feed intake in piglets, enrichment of the environment with foraging substrates seems effective. Enrichment during lactation reduces feed neophobia and results in better pre-weaning growth

performance and higher feed intake during the first 2 critical days after weaning (Oostindjer *et al.* 2010b). Also the facilitation of the transfer of information from the sow to her piglets on what and where to eat is important for her piglets. A loose housed sow during lactation can optimally learn piglets to eat and has been shown to reduce food neophobia and result in higher pre-weaning growth and less damaging behaviours after weaning (Oostindjer *et al.* 2010b). Feed intake of piglets during lactation is stimulated when piglets are able to observe or, even better, participate in feed events of the sow. Moreover, piglets prefer to eat a similar flavoured food and at the same feeder as the sow (Oostindjer *et al.* 2011b). Also the pellet size of the creep feed offered to the piglets affects predominantly post-weaning growth and feed intake. Piglets seem to prefer large pellets (10-12 mm) over smaller pellets (2 mm) which results in earlier onset of feed intake during lactation. Post weaning piglets fed larger pellets during lactation had a 26 % higher body weight gain and 15 % higher feed intake the first 10 days after weaning (Van den Brand *et al.* 2014). Collectively these papers show that stimulation of early solid feed intake during lactation better prepares the piglets to the critical weaning period where resilience is at stake.

## Effects of Current Environment of Challenges in The Environment

The above mentioned examples show that the perinatal environment has profound effects on the resilience of farm animals during environmental challenges and critical transition. However also the current environment of animals can be optimized to make animals cope with challenges in the environment.

As an example, in pigs it has been shown that environmental enrichment imposed after weaning at least partly prevents tail biting (Zonderland *et al.* 2008, Camerlink *et al.* 2015). In a recent study (Dixhoorn *et al.* 2015, In prep.) piglet kept in an enriched housing or barren environment from birth onwards were challenged with a PRRSV (Porcine Reproductive and Respiratory syndrome virus at 14 days after weaning and APP (Actinobacillus Pleuro-Pneumoniae) at day 22 after weaning. The percentage of piglets with APP induced lung lesions (57.1 vs 7.1%) and the extent and severity of lung lesions was substantially lower in enriched housed piglets as compared to barren housed piglets. This suggests that enriched housed piglets are better in coping with multifactorial lung challenges.

Another approach to make animals more resilient in the current environment is to reduce the physiological load of animals during critical transition periods. An example can be found in dairy cattle in early lactation where milk production is high relative to feed intake and animal experience a negative energy balance. Severe negative energy balances in early lactation result in metabolic problems like ketosis and fatty liver syndrome and impair subsequent reproduction. Shortening of the dry period in dairy cattle before onset of lactation has been shown to reduce the negative energy balance in early lactation thereby improving metabolic health and reproduction (Van Knegsel *et al.* 2014, Chen *et al.* 2015). Partly this effect was caused by a lower milk output in early lactation but this was at least partly counterbalanced by extra milk production before onset of the new lactation due to the shorter dry period.

## Conclusion

Health, welfare and productivity in a challenging environment is depended on the adaptive capacity of an animal and the severity of the challenges the animal encounters during its life. The adaptive capacity of animals is shaped by early pre- and postnatal experiences and the actual present environment an animal lives in. Proper preparation in early life and supportive environmental conditions make the animal robust to later life challenges.

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# Genomic Selection for More Sustainable Livestock Production: The French Situation

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## Introduction

The availability of high throughput genomic technologies over the last decade has allowed the development of genomic selection (GS) programs, i.e. the use of genomic information (i.e. DNA chips or sequence data) to genetically evaluate and select candidates to selection. These new tools are likely to change radically breeding programs in both livestock and plant populations. INRA has put GS as one of its research priorities through one of its metaprograms (acronym SelGen - see <http://www.selgen.inra.fr> for details). Over last years, the INRA animal genetics division has devoted a lot of efforts to both theoretical and practical aspects of the development of GS in livestock populations. This paper reviews major breakthroughs achieved over this period of time. Advances made in cattle, which has been by far the most impacted species, will be described first. Applications to other livestock species and theoretical developments will then be presented.

## Genomic Selection in Cattle

The development of GS in French dairy cattle populations has strongly benefited from the implementation of a large-scale (14 chromosome regions, 45 microsatellites) Marker Assisted Selection (MAS) program in Holstein, Normande and Montbéliarde breeds in the early 2000's. After 7 years of activity, more than 70,000 animals had been genotyped, and the efficiency of the MAS program was close to its expectation (Guillaume *et al.* 2008). This large reference population, as well as the experience gained from this first-generation program, were very useful for the development of GS. Indeed, as soon as the BovineSNP50™ beadchip (Illumina Inc, San Diego, USA) has become available in late 2007, it has been intensively used to upgrade the MAS program, leading to a first release of genomic breeding values (GEBV) as early as fall 2008 (Boichard *et al.* 2012b).

GEBV became an official genetic evaluation method in 2009, which allowed genomically evaluated bulls without progeny test to be marketed. An original genetic evaluation model including QTL traced with haplotypes as well as a polygenic residual component was used. QTL were initially detected through GWAS and then by ElasticNet, the latter approach allowing for a larger proportion of genetic variance to be explained (Croiseau *et al.* 2011).

To increase the reliability of GEBVs, a European collaboration resulted in the creation of the Eurogenomics consortium, which gathered research industry players from France, Netherlands, Germany and Nordic countries (followed later by Poland and Spain). Through this alliance, a large reference population of 16,000 Holstein bulls was assembled in 2009, reaching 30,000 in 2015. The gain in reliability was estimated at about 10% for each trait in all countries (Lund *et al.* 2011). This initiative then appeared as an excellent platform for common data management and standardization, for shared R&D - e.g. imputation (Schrooten *et al.* 2014) or use of sequence data -, and for common chip ordering. In 2013, a similar international cooperation was set up in the Brown Swiss breed, a small population in France but a large one worldwide, leading to an official evaluation in this breed in 2014.

Possibilities to build an across-breed genomic evaluation were investigated in the frame of the GEMBAL project funded by the French National Research Agency (ANR) and the French cattle industry. It was based on the use of the High Density chip (HD, 777k), a device INRA contributed to develop in 2010. The goal was to extend genomic evaluations to all beef and dairy breeds by using as much as possible a common reference population. Over 5,000 bulls from 18 breeds were genotyped with the HD chip. The first step of the project, the imputation from 50k to 777k was found to be highly accurate, provided that at least 200 important bulls were genotyped with the HD chip (Hoze *et al.* 2013). Imputation accuracy slightly varied according to breed effective size and genetic relationship between individuals. The second step, i.e.

genomic prediction, gave less favourable results: in agreement with other initiatives at the international level, prediction accuracy only slightly increased when using across breed information, even with INRA model explicitly taking into account QTL effects (Hoze *et al.* 2014). Several reasons can be put forward to explain this somewhat disappointing result: a low proportion of shared QTL between breeds, varying QTL effects according to the genetic background, or more likely insufficient linkage disequilibrium. As a consequence, the idea of an across breed evaluation based on a HD chip was abandoned, but should be revisited in the future using sequence data.

Indeed, sequence data include information on causal mutations, and we can assume that including this information in the evaluation will improve the accuracy, and particularly the persistency, of genomic prediction. However, the huge number of polymorphisms included in sequence data generates a lot of noise, so that an efficient marker selection step has to be developed in order to filter out the vast majority of useless information. This question was addressed through the GENSSEQ project funded by INRA SelGen metaprogram. About 300 whole genome sequences were obtained through different projects in a variety of breeds. Most of them are shared or are going to be shared in the “1000 bull genomes” consortium (Daetwyler *et al.* 2014). In a joint work with Danish colleagues, it was demonstrated that GS accuracy is improved when causative variants or very close polymorphisms (<1kb) are included in the analysis, whereas more distant markers generate noise and are detrimental for the quality of predictions (van den Berg *et al.* 2014). A large-scale project based on GWAS analysis on imputed sequences to identify many causal variants and use them in predictions is currently being developed.

A large-scale use of GS is highly strategic. It is a profitable innovation for farmers with strong consequences on production systems. It is also of major importance for the replacement of reference populations, because the future number of precisely evaluated bulls will be limited (Boichard *et al.* 2015). However, this large-scale use of GS is highly dependent on its cost. In order to reduce genotyping costs, INRA developed in 2011-2012, in the frame of an international consortium gathering USDA, DPI Australia and Illumina, a low-density chip (7K) optimized for imputation over a large range of cattle breeds (Boichard *et al.* 2012a). This chip is now widely used worldwide. In 2012, an automatic procedure was implemented to deliver GEBVs on a weekly basis. In 2013, the chip was improved by adding two types of markers: a) generic markers to further improve imputation or predict targeted genes, b) candidate mutations (5,000 in its fall 2014 V4 release) for large scale validation and, ultimately, to improve genomic predictions. In early 2015, more than 400,000 animals had been genotyped so far, about 80,000 genotyped females had phenotypes and were included in the genomic predictions.

The impact of GS on dairy cattle breeding schemes has been very strong. As soon as 2009, following INRA recommendations, the progeny test of young bulls was stopped and the market share of genomically evaluated bulls increased from 25% in 2010 to 75% in 2014. To limit the increase in inbreeding due to the reduced generation interval, the use of a large number of young bulls as cow and as bull sires was recommended (Colleau *et al.* 2009; Colleau *et al.* 2015) and has been well followed by the breeding companies. The higher efficiency of GS allowed breeding goals to be modified in 2012 towards a higher sustainability by decreasing the weights of production traits and increasing those of functional traits. Several projects are currently developed to implement genomic evaluation for new traits (health, carcass traits, cheese-making properties) and include them in the breeding goal and to set up genomic evaluations for additional breeds.

## Genomic Selection in other Livestock Species

The utility of GS in other livestock species was investigated using a combination of theoretical and practical approaches. In dairy sheep, genomic predictions using Single Step BLUP proved to be more accurate than traditional indices in the Lacaune and in the Basco-Béarnaise breeds (Duchemin *et al.* 2012; Baloché *et al.* 2014). When reference populations are small (e.g. in the Saanen and Alpine goat breeds, in French “Manech Tête Noire” and in Spanish “Latxa Cara Negra Navarra” sheep breeds), multiracial approaches based on the two-step GBLUP or Single Step BLUP appeared as very efficient, with gains depending on the trait considered (Carillier *et al.* 2013; Legarra Albizu *et al.* 2014).

The UTOPIGE project, financed by ANR and the pig and poultry industries, was launched to assess the utility of GS in pigs and in laying hens, where commercial animals are issued from complex crossbreeding plans between several pure lines, with important environmental differences between nucleus and commercial herds. First results appear as promising, with a noticeable gain in accuracy of GEBV as compared to usual

indices in laying hens (Chapuis *et al.* 2015). In horses, GBLUP and Bayes C methods appeared as only slightly more accurate than classical evaluation procedures (Ricard *et al.* 2013).

The interest of GS was also investigated through the modelling of selection programmes. In small ruminants, deterministic approaches were developed to compare scenarios including or not genomic information (different populations or genotyped candidates, maintenance or suppression of progeny testing) and to identify necessary changes in the organisation of selection programmes associated with the use of genomic selection (Shumbusho *et al.* 2013). In pigs, stochastic simulations were used to describe a male line selected for two uncorrelated traits recorded in candidates or their relatives and showed that GS may boost annual genetic trends by 25 to 30% while ensuring a better control of genetic variability (Tribout *et al.* 2012). In all cases, a large reference population and low genotyping costs are necessary. Profitability analyses of these investments are essential, and will allow economically viable solutions to be proposed (Tribout *et al.* 2013).

## Conclusion

GS has markedly changed (dairy) cattle breeding schemes over the last decade, as it appeared as both less expensive and more efficient than traditional selection methods. The situation is less straightforward in other livestock species, but results obtained so far tend to show the interest of an appropriate use of GS in most livestock species. As in cattle, these new tools should be used to improve the sustainability of future livestock production systems.

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# Outcomes of Selection for Residual Feed Intake in Australian Beef Cattle

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## Introduction

One of the largest costs in raising livestock is feed (Archer *et al.* 1999). Thus, genetic improvement should include minimizing feed costs in addition to maximizing production to improve efficiency and profitability. One method of improving efficiency is to select for increased “gross efficiency” or decrease its inverse “feed conversion ratio”. Both of these are simple ratios of weight gain and feed eaten. While they work well for growing stock and are common measures used by feedlot, they are likely to lead to increases in cow size which may be undesirable for cattle breeders.

An alternative trait that can be selected is “residual feed intake” (RFI, Koch *et al.* 1963). RFI is calculated from a linear model of feed intake regressed on average weight and weight gain during a feed test. The residuals from the model are RFI and represent animals that, independent of their size and growth rate, have eaten more or less than expected based on the population in the feed test. In Australia selection lines were developed for RFI in Angus cattle using a post-weaning feed test. The aim was to select young animals with the aim of improving both feedlot feed conversion efficiency and reducing maintenance requirements of cows to improve breeder profitability.

The aim of this paper is to report a series of experiments on the RFI selection line cattle after approximately three generations of selection in a range of scenarios. It includes heifers in an intensive pen trial, steers in a feedlot and cows on pasture over multiple parities. The work represents a large investment by Meat and Livestock Australia and a large team of scientists from 4 state governments and 3 universities.

## Materials and Methods

High and Low-RFI selection lines have been developed at Trangie Agricultural Centre by NSW Department of Primary Industries. Heifers (8 High and 8 Low) aged 6-18 months were fed the same diet at either 105% or 180% of maintenance representing restricted and *ad libitum* intakes. A range of factors were measured with a focus on body composition and metabolism in addition to weight and feed intake.

Steers (68 High and 68 Low) were sent to a commercial feedlot and fed *ad libitum* for 251 days with individual performance including extensive measures of carcass quality (Herd *et al.* 2015). Feed intake was measured at the pen level.

Heifers (81 High and 81 Low) were after their post-weaning RFI test were sent to two research farms, Struan owned by the South Australian Research and Development Institute and Vasse owned by the Western Australian Department of Agriculture and Food (Pitchford *et al.* 2015). After mating they were assigned to either High or Low-Nutrition where the aim was to achieve a 20% difference in feed intake based on stocking rate, not feed quality. They remained in these treatment groups in 2 lines x 2 nutrition x 4-5 replicates = 18 groups for four years where they had the opportunity to raise three calves (18 x 3 = 54 data points). Individual cow weight (monthly), fat and muscle (3 months, Accioly *et al.* 2015) measurements were conducted regularly, calf birth and weaning weights were recorded and feed intake was recorded weekly for the replicate group (not individual, Hebart *et al.* 2015). The High and Low-RFI lines were managed the same to avoid confounding of results.

The economic analysis of the lines modelled a system where cows were pregnancy tested at weaning time each year (Anderton *et al.* 2015). All cows that were not pregnant or did not raise a calf were sold and cows were also sold after raising their 6<sup>th</sup> calf at almost 8 years of age. There was a cost placed on pasture allowing for variation in quality (\$0.0035/MJ = \$40/tonne at 11.5 MJ ME / kg DM) and supplementary hay (\$0.016/MJ = \$140/tonne at 9 MJ ME / kg DM) and returns on weaner calves (\$1.90/kg liveweight), cull heifers (\$1.50/kg) and cull cows (\$1.30/kg). The modelling was conducted in Australian dollars, but at the time the dollar was approximately at parity with the US dollar and so can be considered equivalent.

## Results

Under restricted feeding (105% maintenance) there was no difference in feed intake between the High and Low-RFI heifers (Figure 1) but all of the 6.4% difference in feed intake between the lines on high intake (180% maintenance) was associated with differences in fatness (Lines *et al.* 2015).

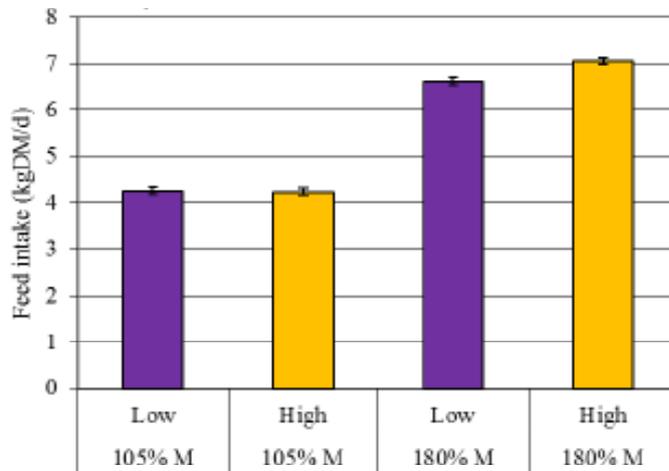


Figure 1. Feed intake of High and Low-RFI heifers at two levels of feeding

The cattle ate 6.87 kg/d on the high intake diet and 39% less (4.20 kg/d) on the low quantity diet. The growth rate on the high diet was reduced to half (0.89 vs 0.44 kg/d). Differences between RFI lines in RFI were significant on the high diet (0.34 kg/d) but not on the low diet where the difference between lines was reduced to almost zero (0.01 kg/d). The change in variation and negligible correlation between diets is presented in Figure 2.

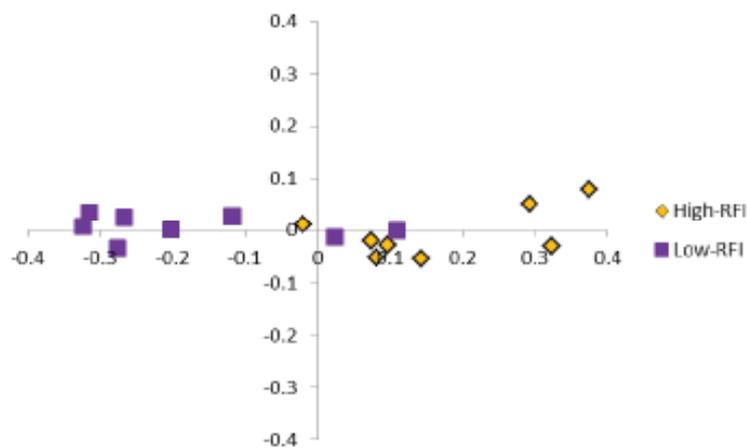


Figure 2. Residual feed intake (kg/d) of heifers at two levels of feeding

The Low-RFI steers were similar weight at the start but grew faster and ate less resulting in a 10% superior feed conversion (Table 1). However, they gained less fat which, depending on the market, could be regarded as a positive or negative. When energy retention was compared to energy intake, half of the 10% difference in FCR was associated with differences in fat deposition. This result is similar to that in the heifer pen trial, although less extreme. The difference in subcutaneous fat depth was greater than that for intramuscular fat.

In order to compare the RFI lines, when an animal in either group dropped body condition to 2 on a 5-point scale (Graham 2006), then animals in both groups were fed supplementary hay. It was always a Low-RFI animal that triggered this supplementary feeding event so the result is that the High-RFI animals were always approximately 2mm fatter than Low-RFI. Cubic splines were fitted to the data as presented in Figure 3 (Accioly *et al.* 2015).

Table 1. Feedlot performance of High and Low-RFI steers

	High-RFI	Low-RFI	Difference (%)
Feed intake, kg/d	11.1	10.4	-6
Gain, kg/d	1.07	1.11	4
Feed conversion, kg/kg <sup>A</sup>	10.4	9.4	-10
RFI, kg/d <sup>A</sup>	0.2	-0.8	-9
Start wt, kg	432	435	1
Slaughter wt, kg	701	714	2
Carcass wt, kg	406	417	3
Dressing %	58.0	58.5	0
Marbling score	3.0	3.0	0
Rib fat depth, mm	21	16	-24

<sup>A</sup>Intake traits measured at pen level and so standard errors cannot be calculated and formal tests of significance are not possible

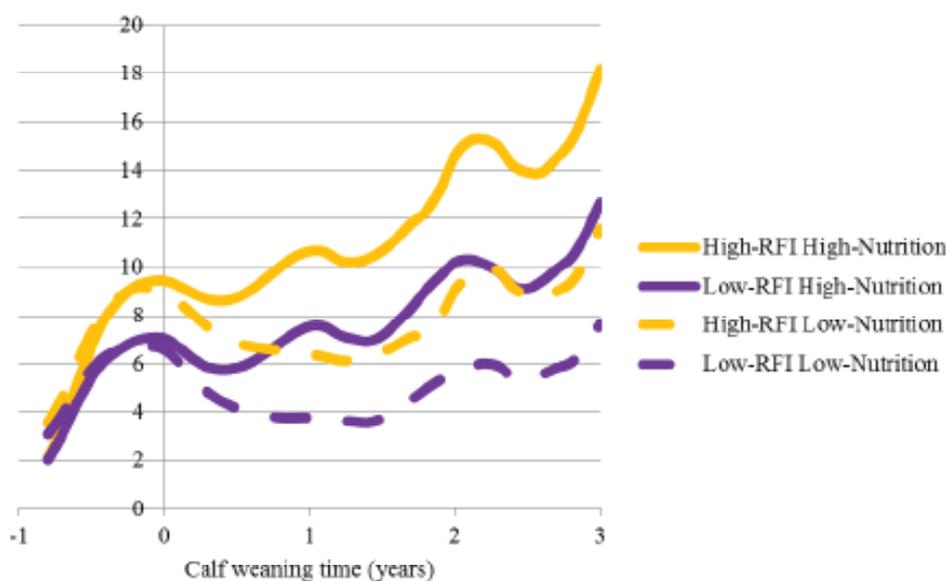


Figure 3. Rib fat depth (mm) of High and Low-RFI cows during multiple parities <sup>A</sup>

<sup>A</sup>Nutrition treatments started after joining (approximately 15 months of age, slightly older at Struan) when heifers were almost 2 years of age. Cows calved soon after this and lactated for 7 months. The data presented is for those cows that weaned a calf at all three opportunities.

On both High and Low-Nutrition, selection for Low-RFI resulted in cows that ate 6% less and had 19% less fat depth, but they had superior residual feed intake (RMEI, Table 2). On Low-Nutrition they also had 7% better maternal productivity when defined as output of calf per energy intake by the cow and calf combined.

The Low-RFI were approximately 5% heavier at weaning and as cull cows. Thus, their income was almost 5% greater. However, there was smaller differences between lines in feed costs so the Low-RFI had superior gross margin on both High (4%) and Low-Nutrition (13%).

## Discussion

The results demonstrate that divergent selection for post-weaning residual feed intake (RFI) has clearly led to improvements in efficiency and profitability in both feedlot finished steers and cows on pasture. Furthermore, the reduced intake has been associated with reduced fatness. In a production system with surplus energy like a feedlot or cows with low stocking rates on good quality pasture, this reduced fatness is likely to be an advantage due to the high cost of depositing fat which is often trimmed off carcasses and sold for a reduced price relative to lean meat.

Table 2. Productivity of High and Low-RFI cows

	High-RFI High-Nutrition	Low-RFI High-Nutrition	High-RFI Low-Nutrition	Low-RFI Low-Nutrition
<i>Efficiency analysis<sup>A</sup></i>				
Weaning rate (%)	84±3	82±3	89±4	79±4
Weaning wt (kg/cow)	226±4	217±4	204±5	197±5
Feed intake (MJ/d)	155±4	149±5	127±5	117±5
Rib fat at mating (mm)	10.5±0.7	8.1±0.7	8.3±0.7	7.1±0.8
Maternal prod. (g/MJ)	4.15±0.16	4.14±0.16	4.49±0.17	4.81±0.17
RMEI (MJ/d)	14.9±4.1	1.2±4.1	4.6±4.5	-9.1±4.6
<i>Economic analysis<sup>B</sup></i>				
Heifer reproduction (%)	73	80	78	70
Cow reproduction (%)	90	81	96	88
Weaner heifers retained	266	287	229	283
Weaner steer weight (kg)	267	283	246	258
Cull cow weight (kg)	671	715	592	610
Income (\$/cow)	556	582	504	529
Expenses (\$/cow)	363	379	298	297
Gross margin (\$/cow)	194	202	206	232

<sup>A</sup>Based on no culling for reproduction averaged over first 3 parities and replicates are groups not individuals. Maternal productivity is weight of calf weaned per metabolisable energy consumed. RMEI is residual metabolisable energy intake adjusted for reproduction (weaning rate and age of calf), cow metabolic mid-weight, cow weight change and weight of calf weaned.

<sup>B</sup>Modelling production system with 1000 pregnant cows at start of production year that culls cows that fail to get pregnant, lactate and after they have raised their 6<sup>th</sup> calf. Calculations done on treatment means so not possible to calculate standard errors. Heifer reproduction is proportion of heifers that got in calf, raised a calf and successfully get pregnant again while lactating. Cow reproduction is proportion of cows that were pregnant, are lactating and successfully get pregnant again while lactating.

As demonstrated in Figure 3, the High-RFI cows were always fatter when given the same amount of supplementary feed as the Low-RFI cows. However, it was the Low-RFI cows that always triggered the initiation of supplementary feeding. If the High-RFI cows were not fed until they reached the same trigger condition (score 2), there would be days of supplementary feeding saved. In the project herein, supplementary feed was given during the dry season to non-lactating cows. They consumed approximately 100 MJ/d during this period (Hebart *et al.* 2015), representing a cost of \$1.60/d. Thus, the difference in gross margin per cow of \$26 would be completely eroded if the Low-RFI cows required 16 days longer supplementary feed. Given the rates of loss of condition during this period (Figure 3), a difference of 16 days could easily have occurred if the lines were managed by their own condition.

The lack of variation in RFI on low levels of feeding in the heifer pen trial (Figures 1, 2) and zero correlation with RFI closer to *ad libitum*, indicates that the result of selection for RFI rather than lower maintenance costs of cows, has altered the appetite of cows. The implication here is that the Low-RFI cattle have a lower drive to eat and so do not gain as much fat as High-RFI cattle. While fat is costly, in production systems as in southern Australia with seasonal pasture availability, it is preferable for cows to eat as much as possible in when the feed is available and this sets them up with additional energy reserves for times of feed shortage. While it is not smart to select for cattle that eat more, it is smart to select for cattle to maximize productivity and these are likely to have greater feed intake. The conclusion from this work matches exactly that by Berry and Crowley (2013) who concluded that ‘the “saved” energy in more efficient animals is likely to have implications for the animal, and this can be best identified by aggressive selection for the phenotype under investigation and evaluating the impact in contrasting production systems.’

## Conclusion

Australian scientists have worked on selecting for feed efficiency in beef cattle for over 20 years. It is heritable and genetic progress has been made. However, it is recommended that the focus of future work should be on traits that are easier to measure such as cow fertility and body condition, calf growth and carcass quality. This is especially the case in the tropics where cattle are raised on lower energy feeds and adaptability traits must be considered.

## Acknowledgement

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# Molecular Mechanisms Regulating Beef Quality in Korean Cattle

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## Abstract

*Intramuscular fat (IMF: marbling) in the longissimus dorsi muscle (LM) of cattle is an important component of traits that influence beef quality. We have conducted several studies in order to understand molecular mechanisms responsible for regulating IMF deposition in beef. In study with Korean cattle steers, our results reveal that the combined effects of increases in lipogenesis, fatty acid uptake, fatty acid esterification, and of decreases in lipolysis and fatty acid oxidation contribute to increasing IMF deposition. Castration of bulls significantly increases IMF deposition. Expression levels of lipid metabolism genes were compared between bulls and steers by real-time PCR or microarray analysis and by Western analysis in various tissues, including LM, fat and liver tissues. Our study shows that castration contributes to increases in lipid uptake and lipogenesis and a decrease in lipolysis in LM tissues, resulting in improved marbling. Our microarray analysis demonstrates that castration alters the transcriptome associated with lipid/energy metabolism, favoring IMF deposition in the LM. We examined whether castration affects adipose cellularity and lipid metabolism gene expression in various fat depots. Our results demonstrate that combined effects of higher adipogenesis, lipogenesis, and cellular fatty acid transport are responsible for the largest abdominal and smallest IMF cells among the fat depots examined. Expression levels of lipid metabolism genes were also compared in the liver between Korean bulls and steers. Hepatic lipid metabolism, including triglyceride synthesis, fatty acid oxidation, and very low density lipoprotein secretion, was not significantly altered by castration. Overall, our study demonstrates that lipid metabolism in the LM and fat depots is important for regulation IMF deposition, whereas that hepatic lipid metabolism has minor effect on IMF deposition. Results of our studies can be applicable to fine nutritional or genetic methods for production of high-quality beef.*

*Keywords: beef quality, fat depot, intramuscular fat, Korean cattle, lipid metabolism, LM, liver*

## Introduction

Intramuscular fat (IMF: marbling) in the *longissimus dorsi* muscle (LM) of cattle is an important component of traits that influence beef quality. We have conducted several studies in order to understand molecular mechanisms responsible for regulating IMF deposition in beef. Gene expression profiles were compared between bulls and castrations in various peripheral tissues, including LM, fat, and liver of Korean cattle, by using functional genomics tools such as a customized bovine microarray.

## Materials and Methods

### Animals and Tissue Collection

LM, various fat tissues, including intramuscular, subcutaneous, abdominal, and perirenal fat, and liver tissues were collected from Korean cattle bulls and steers (Bong *et al.* 2012; Jeong *et al.* 2012). Castration has significantly increased IMF contents, and steer LM meat in these studies had a 3.7-fold higher IMF content than that from bulls

### Gene Expression Analyses Using Functional Genomics Tools

Microarray hybridization, real-time PCR, or Western analysis was used as described ((Jeong *et al.* 2013a).

## Results and Discussion

### A Study with Korean Cattle Steers

In Korean cattle steer group, carcass characteristics and gene expression levels by real-time PCR analysis were measured and the correlation coefficient between IMF content and expression levels of genes for fat deposition and fat removal pathway was analyzed. Results revealed that the combined effects of increases in lipogenesis, fatty acid uptake, fatty acid esterification, and of decreases in lipolysis and fatty acid oxidation contribute to increasing IMF deposition (Jeong *et al.* 2012).

### Comparison of Gene Expression Profiles between Bulls and Steers in The LM of Korean Cattle

Castration of bulls significantly increases IMF deposition. Expression levels of lipid metabolism genes were compared between bulls and steers by real-time PCR, microarray analysis or by Western analysis in LM tissues. Real-time PCR and Western analyses show that castration contributes to increases in lipid uptake and lipogenesis and a decrease in lipolysis in LM tissues, resulting in improved marbling (Bong *et al.* 2012). Our microarray analysis study demonstrates that castration alters the transcriptome associated with lipid/energy metabolism, favoring IMF deposition in the LM ((Jeong *et al.* 2013a). We also found that Wnt/beta-catenin signaling and adipogenic genes are associated with intramuscular fat content in the LM of Korean cattle (Jeong *et al.* 2013b). This study suggests that downregulation of the Wnt/b-catenin signaling pathway genes, but upregulation of Wnt antagonist SFRP4 and adipogenic gene expression following castration, contributes to increased IMF deposition in the LM.

### Comparison of Gene Expression Profiles between Bulls and Steers in The Various Fat Tissues of Korean Cattle

We examined whether castration affects adipose cellularity and lipid metabolism gene expression in various fat depots. Our results show that castration increases body fat cell sizes at various fat depots, and that the up- regulation of adipogenesis and down-regulation of fatty acid  $\beta$ -oxidation may partially contribute to the increased cell size (Baik *et al.* 2014).

### Comparison of Gene Expression Profiles between Bulls and Steers in The Liver Tissues of Korean Cattle

Hepatic expression of genes for lipid metabolism was compared between bulls and steers since the liver has remarkable metabolic flexibility. This study suggests that hepatic lipid metabolism genes involved in triglyceride synthesis, fatty acid oxidation, and very low density lipoprotein secretion was not significantly altered by castration (Baik *et al.* 2015).

## Conclusion

Overall, our studies demonstrate that lipid metabolism in the LM and fat depot is important for regulation of IMF deposition, whereas that hepatic lipid metabolism has minor effect on IMF deposition. Our results could be applicable to fine nutritional or genetic methods for production of high-quality beef.

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# Vaccination and Subclinical Manifestation of Avian Influenza in Indonesia

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## Introduction

Avian Influenza (AI) in Indonesia was observed since July 2003 attacked commercial layer farm in Legok Tangerang West Java and Indonesian government officially declared in January 2004 (Surat Keputusan Menteri Pertanian Republik Indonesia nomor 96/KPTS/PP.620/2/2004 tanggal 3 Februari 2004). Recently AI is endemic among poultry in Indonesia. Vaccination is chosen as one of some strategies to control this disease. Homolog (H5N1) and heterolog vaccines (H5N2 and H5N9) were used in the field for a couple of years (Naipospos 2012). Now only homolog vaccine is allowed to be used in controlling the disease in Indonesia. In Kampung chicken the vaccination is prohibited due to many obstacles in getting the appropriate results of immune response and also the coverage of vaccination was very poor. Unappropriate vaccination caused the subclinical manifestation of this disease. Recently, it is common that highly pathogenic AI virus could be isolated from clinically healthy chicken.

Vaccination of AI succeed to decrease the morbidity and mortality of bird in the field but paralelly to this that the subclinical manifestation of AI among birds was observed especially in the endemic area. The subclinical manifestation of AI caused a serious problem, the spread of disease from area to area is hard to be controlled. The healthy birds may act as a resesvoir of AI virus and play an important role in spreading the virus.

## The Cause of Subclinical Manifestation

The subclinical manifestaion of AI occured might be due to the unappropriate elimination of virus by immune response and cause the shedding of the virus from healthy bird.

1. AI disease is endemic, most of bird produce antibody against AI virus due to the natural infection of AI virus in the field. The variation of virus content in the natural infection cause the variation antibody titers. The host with low titer of antibody mostly expressed the subclinical infection.
2. Unappropriate used of vaccines, the variation of homology between vaccines and field challenge.
3. The changes of surface antigen of AI virus (*antigenic drift* dan *antigenic shift*), and the antibody of vaccine is not match with field challenge (Mahardika *et al.* 2009)



Figure 1. High mortality of layer chicken caused by HPAI subtype H5N1, blue comb, intensive haemorrhagic appearance in many organs. (field cases of AI 2003).

## Character of HPAI subtype H5N1

HPAI subtype H5N1 belonged *Orthomyxoviridae*, RNA virus has . Influenza virus 8 segmented genome. Virus AI is easy to mutate, potential to infect birds, mammals as well as human. HPAI virus has *multiple basic amino acid* (QRERRRKKR//G) in the *cleavage site* of haemagglutinin (Smith *et al.* 2006). The disease mostly acute and spread through contact with contaminated material (Webster *et al.* 1992).

## Vaccination and Subclinical Infection

Vaccination is chosen as one strategy to control AI in Indonesia because the disease has already spread to many provinces in Indonesia. The stamping out measure is impossible to be done. Vaccination is applied in the breeder, commercial layer using homolog (H5N1) as well as heterolog (H5N2, H5N9) killed vaccines.

Vaccination significantly decrease the incidence of AI in the field (Figure 2). The mortality of chicken decrease significantly after vaccination compared to the mortality in the year 2003 even the cases spread widely (Asmara 2007).

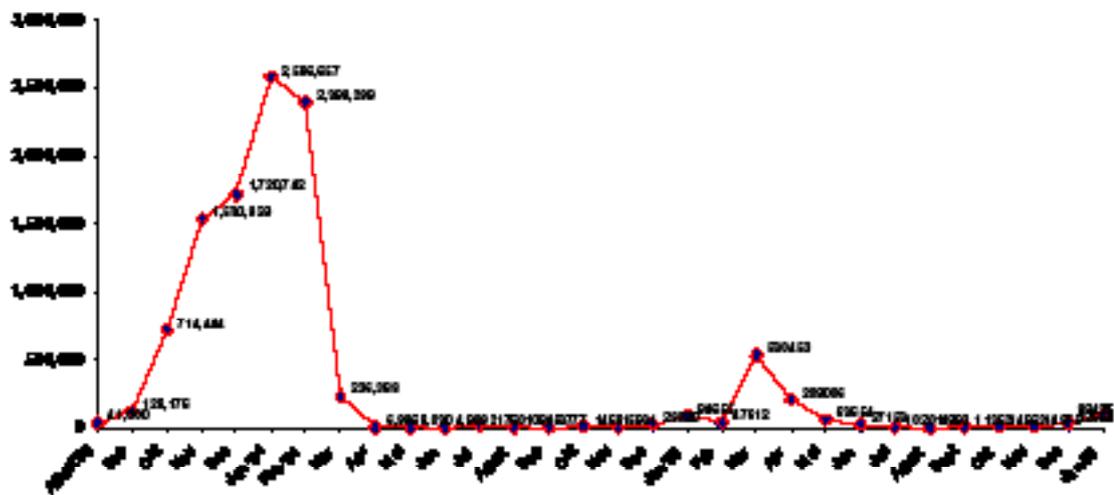


Figure 2. High mortality of chicken caused by AI in the period of September 2003-February 2004, and the mortality of chicken decrease significantly after mass vaccination is applied in February-March 2004 (Ditjennak 2005)

Vaccination of AI induce the protective level of antibody in chicken. One week after the second vaccination (booster) the titer of antibody using HI test higher than 64. The vaccination of AI in backyard chicken is still problematic, the coverage of vaccination is lower than 30% and the protective level of antibody is hard to be reached. This might cause the manifestation of subclinical infection.

## Observation of Subclinical Infection in the Field

The research was done in the period of 2006-2008 and showed that the presence of genetic material of HPAI virus can be detected using RT-PCR among vaccinated as well as non vaccinated backyard chicken with low antibody titer.

Table 2. The presence of HPAI H5N1 among vaccinated and unvaccinated backyard chicken in Banten province (Wibawan *et al.* 2006)

Daerah	Kecamatan	Hasil PCH (jumlah)			
		Vaksinasi		Non-Vaksinasi	
		HI+	HI-	HI+	HI-
Kab. Tangerang	Kronjo			H5 (1)	
	Mauk				H5 (1)
	Paku Haji	H5 (1)		H5 (1)	H5 (2)
Kab. Serang	Kresiek			H5 (1)	H5N1 (1) H5 (1)
	Carenang		H5 (2)	H5 (1)	H5 (1)
	Kragilan			H5 (1)	
Kab. Lebak	Petir	H5 (2)			
	Cibadak	H5 (1)		H5 (1)	H5 (2)
	G Kencana	H5 (1)			H5N1 (1) H5 (1)
Kab. Pandeglang	Muncang	H5 (1)			
	Bayah		H5 (2)		
	Cikeusik			H5N1 (1)	
Kab. Cilegon	Panimbang		H5N1 (1)		
	Jombang		H5 (1)		
	Cilegon		H5N1 (1)		

### Water Fowl as Bio-Indicator of Viral Environment Contamination

Water fowl can be used to justify the AI viral contamination of the environment. The research was done in 2006, in Bogor, Sukabumi, Cirebon and Indramayu (Susanti *et al.* 2007).

Two HPAI virus could be detected among healthy duck in Nagrak Sukabumi using RT-PCR, (NG 26 dan NG 29).

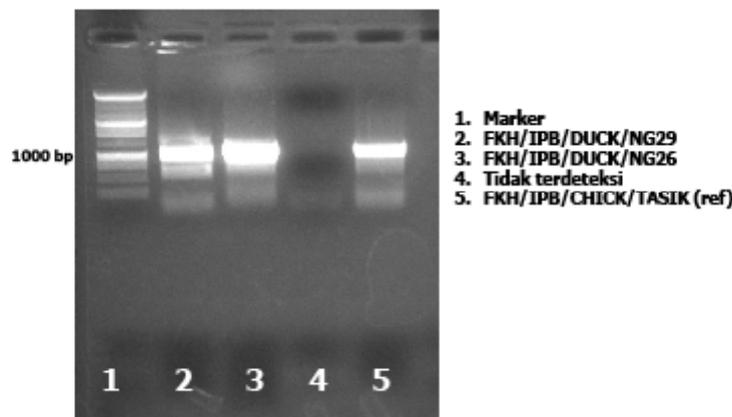


Figure 3. Two H5N1 isolates (NG29 dan NG26) were isolated from healthy ducks in Nagrak Sukabumi (Wibawan *et al.* 2006)

The sequencing of *cleavage site* of haemagglutinin of these isolates (AI FKH/IPB/Duck/NG26 and AI FKH/IPB/Duck/NG29) showed polybasic amino acid codon *RESRRKKRR*. These results confirmed that these 2 AI isolates belonged to HPAI (*highly pathogenic avian influenza*). In the infection experiment using the 2 isolates in the laboratory (BSL-3 Medion) showed that the presence of virus could be detected in the intestinal tract of healthy duck with immunohistochemistry using monoclonal specific antibody (Temasek Life Science Laboratory Singapura (Figure 4). Comparable results was also reported by Webster *et al.* (1978).

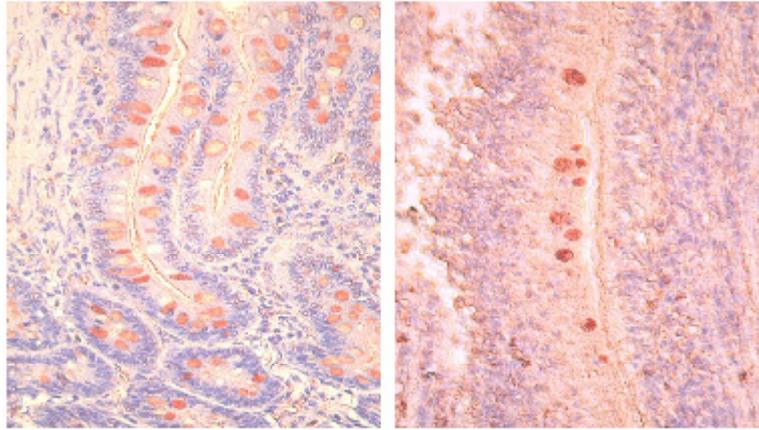


Figure 4. The presence of AI virus H5N1 (FKH/IPB/Duck/NG29 in intestinal mucosal cells of healthy duck in the experiment 2 days after infection) (10x40) ( Wibawan *et al.* 2006b).

These results indicated that the healthy duck might be played an important role as reservoir of AI virus, in spreading of the disease and in contaminating the environment, especially through the faeces. The similar results was reported by Susanti *et al.* (2007, 2008a, 2008b).

## Conclusion

1. Subclinical manifestation of AI was observed among vaccinated and unvaccinated birds in the AI endemic area.
2. The presence of AI specific antibody in the host might play an important role in the expression of subclinical manifestation of AI

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# The Role of Feed Additives in Tropical Animal Farming Industry with Emphasis on Organic Acids

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## Agricultural Market Trends

Global contemporary animal production quadrupled during the past 50 years and totaled in 308mill MT of meat (2013). Asia is the main animal production center and has leading responsibility and contribution to decreasing the world hunger by 2050.

In more detail, the world's population will increase by about 30% to 9.1 billion by 2050 with most of its population in developing countries. Urbanization will continue worldwide and is foreseen to reach 70% by 2050 compared to 49 % today. In order to feed this larger and more urbanized population, global food production needs to increase by about 70% consisting of an annual cereal production rising to about 3 billion tons (from 2.1 billion today) and an annual meat production to 470 million tons, respectively (FAO 2009).

Considering, however, the annually declining growth rates of major cereal crops globally, agriculture is challenged to develop new technologies confronted to compete with natural resources (land, water), climate change and habitat preservation. International trade of feed raw materials and agricultural end consumer products will intensify to ensure food security. To manage these generally outlined challenges, political will combined with professional communication and networking is required. In short, farmers need to produce more from less land with fewer hands. The world compound feed production is approaching to an estimated 1 billion tons annually as to comply with the increasing demand on animal protein in agricultural husbandry (livestock, dairy and aquaculture) (IFIF 2012).

## Feed Additive Markets

Feed additives are food supplements for farmed swine, poultry, ruminants as well as fish and shrimp to control infectious diseases, thereby to support the animals' growth and life performance through a stronger condition and welfare under intensive farming conditions. Feed additives include vitamins, minerals, amino and fatty acids and are classified into nutritional and non-nutritional additives (Figure 1).

The global animal feed additives market was valued at 14.9 billion USD in 2013 and is estimated to reach 20 billion USD, having a compound annual growth rate of 4.2% from 2013 to 2020 due to increasing demand for high nutritional meat available at low costs (Watt 2014). Amino acids gained popularity among meat producers as growth amplifiers to expedite returns of investments under intense farming conditions. Lysine, tryptophan, methionine and threonine are presently the main amino acids used as feed additives. The EU-wide ban of antibiotic growth promoters in animal feeds since 2006 made feed acidifiers the second largest revenue-generating segment followed by vitamins. Antioxidants and feed enzymes are expected to have notable growth during the forecast period though with smaller market size.

The Asia-Pacific region accounts for one of the biggest animal feed additive markets in poultry and swine production driven by increasing per capita meat consumption and EU-export orientation (Marketsandmarkets 2014). For the latter, increasing human awareness towards food safety and high nutritional meat quality with concomitant prevention of animal disease outbreaks are the main factors accelerating the feed additive market growth. Feed additives are incorporated as small dosages into the basic feed mix catalyzing daily animal weight gain through enhanced digestibility and feed conversion with more efficient nutrient uptake. Gut health is improved by decreasing pathogen but increasing beneficial bacteria with parallel prevention of digestive tract infections. Thus, strong improvements of meat quality for human consumption are obtained.

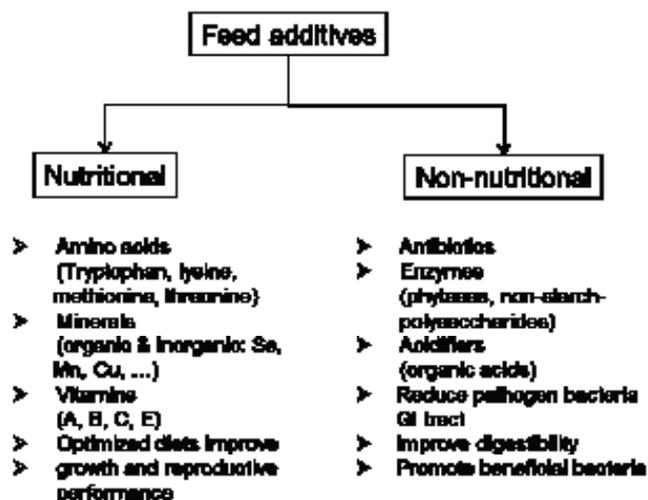


Figure 1. Effects of feed additives in animal nutrition

## Organic Acids in Agricultural Farming

With tropical humidity and temperature climate, pathogen multiplication is exponentially high compared to cooler regions. Intense tropical farming conditions lead to vast feed contamination and disease outbreaks on intensely farmed animals followed by growth reduction and mortalities. The increasing use of feed acidifiers is driven by increasing consumer demand for healthy meat products in developed economic and most recently, leading Asian companies export high quality agricultural meat of swine, poultry, fish, and shrimp into the EU. Food security impacts the complete food chain and here, organic acids play a vital role as to replace antibiotic growth promoters.

This scenario underpins the 6% annual growth of the feed acidifier market being projected to reach 2.7 billion USD by 2018 (PRN Newswire 2014). As feed is one of the most expensive components in farming, special care and knowledge is needed to manage animal feeds efficiently.

Organic acids are natural by-products of microbial fermentation being used in agricultural farming for feed preservation ever since; a main reason why the industry took organic acids as the alternative to replace antibiotic growth promoters in animal feeds. At present, acids are the most cost and performance effective option available to the feed industry.

As organic acids alter the gut micro flora ecology of animals, a pathogen contra beneficial bacteria controlling instrument is provided and feed-applied, often in combinations of various organic acids. This group of “acidifiers” is known to boost animal growth performance, gut health integrity and to enhance daily weight gain and feed conversion – a full concept of economic agricultural factors with ecologically sound effects to a healthy animal nutrition for human consumption.

The efficacy of an acidifier in animal feeds depends mainly on the three-factorial concept of either single or multiple blended organic acids or their salts:

- (i) the organic acids’ molecular weight
- (ii) their minimum inhibitory concentration (MIC) to become bacteriostatic or bactericidal and
- (iii) their recommended dosage in animal feeds (Figure 2).

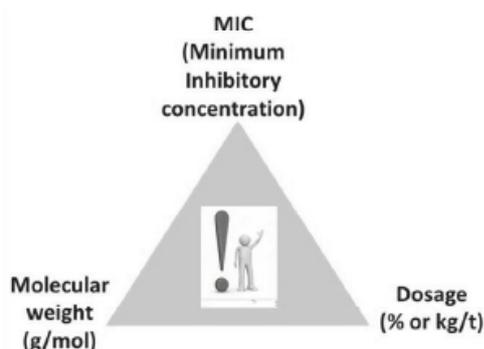


Figure 2. The three-factorial concept of acidifier application into animal feeds

Here, agricultural acidifiers mixed onto feeds vary in terms of organic acid combination to fulfill specific actions in feed raw materials, feed mill hygiene programs or finished feeds ingested and digested by the animal.

## Mode of Action of Organic Acids Against Organotrophic Bacteria

Effects of organic acids used to counteract pathogen bacteria inside the digestive tract of farmed animals vary depending on their dissociation constant and are expressed as pKa value for each acid. This pKa value defines the pH each organic acid is in equilibrium of 50% dissociation and 50% un-dissociation. Thus, the lower the pK value, the stronger the acid. Acids used as feed additives have pKa values between 3 and 5 (Table 1). Further, their molecular weight and MIC are measures to reduce pH and antimicrobial activity.

Table 1. Molecular weight and dissociation constants (pK<sub>a</sub>) of organic acids well accepted for animal feeds and drinking water application to reduce pathogen bacteria residing in digestive tracts of intensively farmed animals

Chemical formula	Organic acid	Molecular weight [g mol <sup>-1</sup> ]	pK <sub>a</sub> value
CH <sub>2</sub> O <sub>2</sub>	Formic acid	46.03	3.75
C <sub>2</sub> H <sub>4</sub> O <sub>2</sub>	Acetic acid	60.05	4.76
C <sub>3</sub> H <sub>6</sub> O <sub>2</sub>	Propionic acid	74.08	4.86
C <sub>3</sub> H <sub>6</sub> O <sub>3</sub>	Lactic acid	90.08	3.08
C <sub>4</sub> H <sub>8</sub> O <sub>2</sub>	Butyric acid	88.12	3.82
C <sub>6</sub> H <sub>8</sub> O <sub>7</sub>	Citric acid	192.1	3.14
C <sub>6</sub> H <sub>8</sub> O <sub>2</sub>	Sorbic acid	112.13	4.76
C <sub>7</sub> H <sub>6</sub> O <sub>2</sub>	Benzoic acid	122.12	4.19

Applying the three-factorial concept of molecular weight, minimum inhibitory concentration and dosage of acidifiers into animal drinking water or feeds, pathogen bacteria residing inside the farmed animals' (poultry, swine) digestive tract can drastically be reduced or eliminated. This likewise holds true for aquaculture farmed fish or shrimps.

Table 2. Minimum inhibitory concentrations (MIC) determined based on pure organic acids against pathogen bacteria (Strauss & Hayler 2001, Skrivanova 2006, Pundir & Jain 2011, Sava 2011)

Chemical formula	Organic acid	Minimum inhibitory concentration (MIC) [%]								
		Pathogen bacteria*								
		A	B	C	D	E	F	G	H	I
CH <sub>2</sub> O <sub>2</sub>	Formic acid	0.10	0.13	0.10	0.10	0.13	0.10	0.10	0.13	0.50
C <sub>2</sub> H <sub>4</sub> O <sub>2</sub>	Acetic acid	0.10	0.10	ns	ns	ns	ns	ns	0.5	0.50
C <sub>3</sub> H <sub>6</sub> O <sub>2</sub>	Propionic acid	0.13	0.20	0.20	0.20	0.23	0.23	0.20	0.23	0.25
C <sub>3</sub> H <sub>6</sub> O <sub>3</sub>	Lactic acid	0.30	0.40	0.23	0.30	0.30	0.30	0.30	0.40	ns
C <sub>4</sub> H <sub>8</sub> O <sub>2</sub>	Butyric acid	ns	ns	ns	ns	ns	ns	ns	ns	0.45
C <sub>6</sub> H <sub>8</sub> O <sub>7</sub>	Citric acid	ns	ns	ns	ns	ns	0.40	ns	ns	ns
C <sub>6</sub> H <sub>8</sub> O <sub>2</sub>	Sorbic acid	ns	ns	ns	ns	ns	ns	ns	ns	0.45
C <sub>7</sub> H <sub>6</sub> O <sub>2</sub>	Benzoic acid	ns	0.10	0.10	ns	ns	ns	ns	ns	0.50

\* A – Salmonella typhimurium, B – Escherichia coli, C – Listeria monocytogenes, D – Campylobacter jejuni, E – Clostridium botulinum, F – Clostridium perfringens, G – Pseudomonas aeruginosa, H – Staphylococcus aureus, I – Aspergillus flavus; n.s. – not specified

As all organic carboxylic acids are weak acids, their dissociation in organic solvent like bacteria cell plasma is based on the Le Châtelier principle in releasing protons with concomitant pH reduction. The most efficient organic acids are those with the smallest molecular mass and lowest MIC, as the highest number of organic acid molecules will be able to enter into, e.g. the gram-negative bacteria, thereby most efficiently reducing its pH-value with the highest ability to kill.

Organotrophic bacteria obtain their nutrient and energy sources for living, growth and reproduction from organic media, such as the carbon atom of organic acids. As they are most vital within neutral pH-ranges, the digestive system of animals, however, works within acidic pH-ranges, organic acids incorporated into animal feeds and ingested by the animal can kill pathogen bacteria in the digestive tract by the bacteria's pH reduction in several ways.

For gram-negative bacteria, organic un-dissociated acids reaching near such bacteria can diffuse through its cell wall as the organic acid is being perceived as a nutrient (Figure 3).

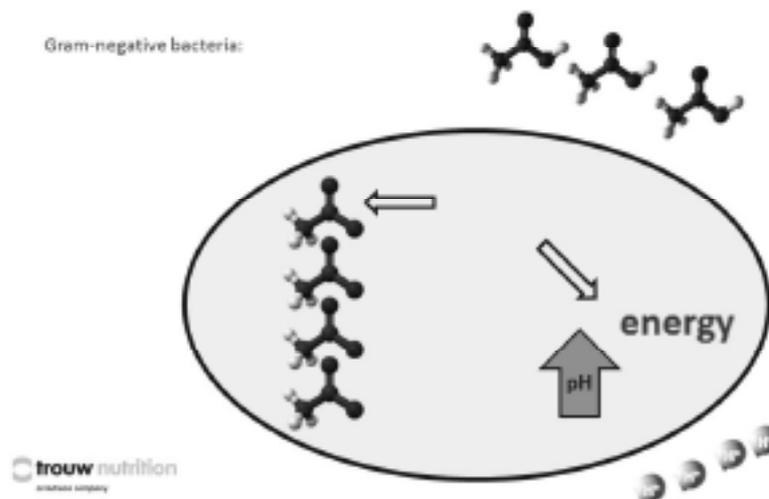


Figure 3. Mode of action of organic acids entering into gram-negative bacteria

Once dissociated inside the bacteria, its pH will shift towards acidic triggering the bacteria to release energy in order to neutralize the pH back to its favorable conditions. As with the feed ingested, more and more organic acid molecules will reach and eventually penetrate and dissociate inside gram-negative bacteria, less and less energy resources can be released. The bacteria will weaken and finally die.

## Mold Prevention in Animal Feeds

The potential of organic acids as forage and grain preservative has a fundamental contribution to feed hygiene by suppressing the growth of molds, yeasts and bacterial pathogens, thus allowing more efficient use of feed resources. Using organic acids in feed preservation has been accepted globally in the agricultural sectors as feed raw materials and finished feeds will result in higher nutritive quality and thereby giving better health conditions and growth to farmed animals leading to higher economic efficiency.

However, due to humid weather with high temperatures or tropical monsoon rains, huge amounts of feed raw materials rot away due to often insufficiently managed or available storage conditions. The global food waste amounted worldwide to 1.3 billion tons in 2013; an equivalent to one third of the annual global food production (FAO 2013) causing severe economic losses and environmental harm. Additionally, harvested decayed grains infest new supplies, intensifying mold and mycotoxin development. About 54% of global food waste occurs during production, post-harvest handling and storage contributing to about 870 million people daily staying hungry. While the annual grain wastage in Australia is estimated to account for only 0.75%, 3.2 million tons (16%) had been recorded for Pakistan. India wastes annually 21 million tons of wheat, which is equivalent to the entire annual wheat production of Australia. Considering the FAO strategic goal adopted at the 1996 World Food Summit to eliminate world hunger by 2050 in developing countries, an even only 10% decrease of global feed raw material waste through professionally engineered storage facilities would picture a significant contribution.

To prevent molds and mycotoxins in feeds causing weak animals ill of metabolic or reproductive diseases and insufficient growth performance, non-corrosive preservative products easy to handle, yet guaranteeing highest grain preservation are needed. Propionic acid combined with benzoates and propionates found wide market acceptance as likewise propionic acid mixed with its buffered propionates and weak organic (e.g. formic, acetic, lactic) acids. Silo-stored but preserved corn ranging from 14 to 30% moisture content is unlikely to mold over several months. Additionally, the free surface water as one main source for mold development is being bound through surfactants guaranteeing safe feed raw materials processed to high value finished feeds for best animal performance. Aside from strong preserving non-corrosive products, leading industry players additionally emphasize on computerized feed raw material and finished feeds service programs including laboratory analysis and automated feed moisture management. Feed mills will be enabled to store and manufacture their products with outstanding quality parameters for best animal

feed production ahead of competition. Further, feed preservation with organic acids reduces energy costs compared to conventional methods (electric cooling or drying).

## Pathogen Bacteria Inhibition in Animal Digestive Tracts

Organic acids incorporated into feeds ingested by animals show several advantages (Freitag 2007):

- (i) Feed acidification and thereby less pathogen bacteria contamination
- (ii) pH optimization in stomach and small intestine and
- (iii) increased feed digestibility and nutrient uptake
- (iv) inhibition of pathogen microbial growth in the intestine

Often, acidifiers are argued to reduce intestinal pH, which however would refer to an empty digestive tract. As animal feed is alkaline, added acidifiers will support reducing the animals' digestive juice production in the upper digestive tract (stomach) and thereby supporting its energy metabolism for converting such saved energy into growth. Protein digestion is improved by triggering pepsinogen to produce more pepsin due to acidifiers intensifying the conversion of food proteins into peptides. Likewise, mineral digestion will be increased. In the lower digestive tract pancreatic juices will increase as buffered organic acids dissociate to more intensely contribute to nutrient digestion and uptake through the gut. Here, dissociated organic acids lower the pH in the bacterial environment and inhibit growth of intra-luminal pathogens. Undissociated organic acids additionally can penetrate gram-negative bacteria to reduce its pH to unfavorable conditions triggering cell death. Both effects are beneficial for beneficial bacteria which are increasing as pathogen bacteria are being reduced (eubiosis). Often, acidifiers are combined of short and medium chain fatty acids (e.g. triglycerides such as palm kernel oil) as the latter are able to penetrate also gram-positive pathogens. Once this cell wall is opened, the short chain fatty acids (e.g. formic, acetic, propionic acids) can enter and kill bacteria cells through pH-reduction and DNA disruption.

In swine farming, sows are strongly conditioned during farrowing as acidifiers are recommended some weeks before farrowing until weaning. Several studies show and all over stronger condition of sows in terms of less back fat loss, higher appetite during the energy-consuming period of farrowing and additionally, are giving the piglet a stronger start-up phase as their digestive tract is still less developed. Thus, post-weaning diarrhea reduction, better weight gain and less mortality occur under rural and tropical farming conditions (Lückstädt & Kühlmann 2013, Lückstädt *et al.* 2014). Combining short chain fatty acids (SCFA) with minimum dosages of medium chain fatty acids (MCFA) in various piglet trials resulted in catalyzing effects of SCFA through MCFA underpinning synergistic effects of the two (v Hess & v Gils 2002). A study revealed that piglets receiving common commercial starter feeds including a SCFA blend were given additionally a 0.1% MCFA. This resulted in a 19% of average daily weight gain (ADG) during the first two weeks of trial and continued to be of 7% higher ADG compared to control piglets during the entire 44 day trial.

Likewise, increased live weight, reduced feed consumption, better broiler indices (European Broiler Index, EBI) were obtained for broilers having received acidifiers incorporated into feeds compared to control diets (Lückstädt *et al.* 2011). A study in Thailand (Kühlmann *et al.* 2012) revealed enhanced performance of for hisex brown layers under Asian conditions having receive an acidifier diet compared to control hens. Egg laying performance was improved especially during the later laying period from weeks 36 onwards with a 91% reduction of *E. coli* in feces samples. A combination of organic acids and medium chain fatty acids incorporated into broiler feeds resulted in strong growth of bactiracin, an antimicrobial growth promoter reducing pathogens of *Clostridium perfringens* in broilers (v Dam 2006).

In aquaculture, acidifiers increased in importance applied to feeds for high market value fish (tilapia, Asian seabass) and shrimp (white leg shrimp), especially considering the strong break down of Southeast Asian shrimp production caused by the Early Mortality Syndrome (EMS) in Thailand or Vietnam. Aside from an improved growth performance of white leg shrimp receiving acidifiers in diets, strong mortality reduction of the gram-negative marine luminescent bacteria *Vibrio harveyi* infected shrimp had been obtained (Kühlmann & Lückstädt 2013). Likewise *Vibrio harveyi* challenged seabass having received acidifiers in their diets showed a stronger growth and health performance with reduced mortalities in a university trial in Thailand (Lückstädt & Kühlmann 2014).

## Conclusion

Feed additives have become an essential part of feed manufacturers and animal nutrition for high quality global meat production. Its rapid growth indicates the strong demand and progressive development for a modern agricultural industry to improve the efficiency of animal production, to alleviate environmental pollution, to maintain animal health and welfare and to assure safe food production of best quality.

The incorporation of organic acids and their salts into animal feeds supports animal health and growth performance. Through organic acids, enzyme secretion, nutrient digestibility and retention result in improved feed conversion and daily weight gains. Economic benefits result in lesser feed consumed with faster growth of animals, thereby shortening production cycles. Additionally, pathogen bacteria are reduced in the intestine as organic acid contribute to improved gut health by increased production of beneficial bacteria and animal condition.

Trouw Nutrition as a leading company in the feed additive market is not only selling high quality products, but providing the unique combination of market specific solutions, vividly applicable models and customer tailored services. This is boosting high quality animal feed production and farming in the Asia-Pacific region. Optimizing animal nutrition of high value market players encompasses concepts, products and nutritional know-how in professional responsibility to 'Feeding the Future', the ultimate company goal.

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# Recent Advances in Gut Microbiology Research in Relation to Animal Nutrition

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## Abstract

*Genetic and biological diversity of gut microorganism is an important area of scientific research. Taking ruminant livestock as an example, the inhabitant of the rumen microbial eco-systems consisted of a complex consortium of different microbial groups living in the symbiotic relationship with the host. These two parties act synergistically for the bioconversion of fibre-based feed into volatile fatty acids which serve as a source of energy for the animals. Our current knowledge began with microscopic studies in the 1930s and 1940s of protozoa and bacteria. Broad classes of bacteria were identified from their ability to ferment different substrates, but even with the improved culture techniques developed by Hungate, limitation to get access on the information of the role of individual species in the fermentation process is still notable. The global microbial diversity presents an enormous, largely untapped genetic and biological pool that could be exploited for the recovery of novel genes, biomolecules for metabolic pathways and various valuable products. The use of molecular techniques in rumen or gut studies has confirmed the complexity of the microbial ecosystem and revealed the diversity of the microbial community. Over the past 30 years, researchers studying the gut microbial community have benefited greatly from the development of molecular techniques. Several DNA-based molecular methods have been developed. Enumerating and identifying of microbial populations within the rumen by culturing and microscopic can be time consuming, cumbersome and inconclusive. Introduction of Real-time PCR has become a powerful tool that allows for rapid quantification of a target DNA sequence through the design of specific primer sets. Apart from that, identification of individual species up to strain level can be achieved by analysis based on 16S rRNA gene which has provide extensive information about the taxa and species present in an environment. It is however, data generated from this analysis can only provide us with little information about the functional role of the complex microbes within the community and the genetic information they carries. The rapid development of the new generation sequencing technologies has created tremendous opportunities for genomic studies. Metagenomics is a rapidly growing field of research that focus on genomic study from all environmental microbes as opposed to genome from individual isolated organism from environment and cultured via in vitro approach. Metagenomic study addressed the limitation of information obtain from uncultured microorganisms to understand their diversity, functions, cooperation and evolution in any environment samples and also digestive system of animals and humans.*

*Keywords: gut, genomic, metagenomic, microorganisms*

Genetic and biological diversity of gut microorganism is an important area of scientific research. Taking ruminant livestock as an example, the inhabitant of the rumen microbial eco-systems consisted of a complex consortium of different microbial groups living in the symbiotic relationship with the host. The composition of microorganisms in the gut environment is highly influenced by external factors, such as diet, feeding frequency, geographical location, age, and ruminant-host interaction (Hungate 1966; Dehority & Orpin 1997). Understanding the intricacies of this interaction is necessary that will lead the researchers to evaluate what were considered to be important microbial population within the rumen with respect to fermentation products, fiber-degradation, methanogenesis and bacterial predation (Mackie 1997). Herbivorous animals are either foregut or hindgut fermenters that rely on microbial fermentation within their gut to attain energy from plants. Animal productivity can be improved in many ways, these include optimizing the functioning of existing microbial ecosystems, or manipulating these ecosystems through intervention of modern technology. For that reason, it is necessary to assess the functions on gut microbial population together with the significance of these functions within natural ecosystems in order to optimize productivity.

Our current knowledge began with microscopic studies in the 1930s and 1940s of protozoa and bacteria. Broad classes of bacteria were identified for their ability to ferment different substrates, but even with the

improved culture techniques developed by Hungate (1950) and have been modified in many ways in order to adapt them according to the requirements of certain kind of studies and to the particular needs, but still, limitations to get access to the information about the role of individual species in the fermentation process is still notable. The global microbial diversity presents an enormous, largely untapped genetic and biological pool that could be exploited for the recovery of novel genes, biomolecules for metabolic pathways and various valuable products.

Over the past 30 years, researchers studying the gut microbial community have benefited greatly from the development of molecular techniques. The use of molecular techniques on microbial population in the rumen or gut of animals has revolutionized our understanding of the complexity of the microbial ecosystem and revealed the diversity of the microbial community in that environment. Molecular biology has made significant contributions to the study of gut microbial populations and their functions, allowing specific microbes of interest, for example, predominant fiber-degrading bacteria to be analyzed in a quick, sensitive and accurate manner, regardless if they are culturable or not. (Gregg *et al.* 1996; Kobayashi and Onodera 1999; McSweeney *et al.* 1999). Enumerating and identifying of microbial populations within the rumen by culturing and microscopic can be time consuming, cumbersome and some time is inconclusive. For this reason, several DNA-based molecular methods have been developed. For instance, conventional cloning and hybridization using a specific probe is among the earliest methods used to determine microbial population. However, these techniques do not provide any information on the dynamics of the microbial populations in complex ecosystems and potential effects of environmental changes on such populations. This limitation has been overcome by the introduction of genetic fingerprinting method known as denaturing gradient gel electrophoresis (DGGE). The dynamics of the microbial populations in complex ecosystems and potential effects of environmental changes on such populations can be investigated via this method. A major advantage of this method is its potential to monitor changes occurring in various microbial communities that are undergoing different treatments or modifications via visually profile. It is a rapid and efficient separation technique of same length DNA sequences (amplified by PCR), which may vary as little as a single base pair modification (Muyzer *et al.* 1993; Sheffield *et al.* 1989). Nevertheless, this method offer restricted information on less abundant taxa, which may bias the overall microbial community present in that environment (Castro-Carrera *et al.* 2014).

Study on the microorganisms population shift is necessary when the effect of certain treatment on the population is investigated. For instance, the introduction of Real-time PCR has become a powerful tool that allows for rapid quantification of a target DNA sequence through the design of specific primer sets and in some cases an internal probe. It has been shown to be a robust, highly reproducible and sensitive method to quantitatively tract microorganism population changes under varying environmental or experimental conditions. This method has enabled us to quantify taxonomic or functional genetic markers present within a mixed community from the domain level down to the quantification of individual species or phylotypes (Smith and Osborn 2009). To name a few, the RT-PCR has been successfully applied to samples extracted from rumen content to monitor microbial population changes in the rumen (Tajima *et al.* 2001; Ouwerkerk *et al.* 2002; Klieve *et al.* 2003). The requirement of sequence data of the specific target gene of interest has become a major limitation of RT-PCR. The sequence information has been primarily derived from gene fragment sequences from cultured organisms and/or from clone libraries generated by PCR using primers that based on current sequence knowledge. Therefore, accessing the unculturable microorganisms using this method is inevitably limited to those that have already been characterized (Smith and Osborn 2009).

The perhaps most widely used technique in microbial ecology has been sequencing of environmental clone libraries. The traditional target for such studies has been PCR-amplified of 16S rRNA genes. This approach was the first taken in the early 90s, and has been applied to a large variety of microbial ecosystems ever since (Eckburg *et al.* 2005; McCaig *et al.* 1999; Sanger *et al.* 1977). Invented by Frederick Sanger's in the mid-70s, he has named it as a chain termination method (Sanger *et al.* 1977) that has been the dominant DNA sequencing technology. It is a well-established and a common amplicon sequencing method used to study phylogeny and taxonomy of samples from complex microbes that are difficult or impossible to study. It is, however, data generated from this analysis can only provide us with little information about the functional role of the complex microbes within the community and the genetic information they carry. The rapid development of the new generation sequencing technologies has created tremendous opportunities for genomic studies. The development of techniques allowing for large fragment cloning has set the stage for a new environmental genomics science that are known as metagenomics. The term 'metagenomic' was firstly introduced by Handelsman *et al.* (1998). It has become a rapidly growing field of research that focus

on genomic study of all environmental microbes as opposed to genome from individual isolated organism from the environment and cultured via *in vitro* approach. This approach allows researchers to describe not only the phylogenetic diversity of microbes in natural environments, but also the genetic and metabolic diversity, without the need for prior cultivation (Rondon *et al.* 2000; Tringe *et al.* 2005; Tyson *et al.* 2004). It also addressed the limitation of information generated from uncultured microorganisms to understand their diversity, functions, cooperation and evolution in any environment samples including the digestive system of animals and humans. The implementation of metagenomic has vastly reduced the costs of DNA sequencing, while achieving equally vast increases in throughput (Marguiles *et al.* 2005; Ronaghi *et al.* 1998).

As a conclusion, as our understanding of individual and group of gut microbial population in which they exist has progressed, genes that are responsible in enzyme production by the microbes has been successfully encoded, cloned, characterized and used in order to enhance its function. Furthermore, there is an ongoing need for a search on a wide range of novel genes and enzymes required by the ruminant to improve digestion of fiber in low quality forage, selection of predominant rumen microbes on specific substrate and studying the interaction between nutrient and hosts on the production aspect. By having information about what is there and what have been produced by the microbes, it is hoped that, with available information generated by the molecular studies, the researchers could improve the productivity of the livestock by manipulating the gut microbial population accordingly.

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**THEME A.**  
**ANIMAL PRODUCTION, TECHNOLOGY AND INDUSTRY**

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**Proceeding of the 3<sup>rd</sup> International Seminar on Animal Industry,  
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# Beef Cattle Production System, Constraints and Opportunities for Small Farmers in South Central Timor Regency, West Timor

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## Abstract

*Bali cattle is important in South Central Timor Regency, counted for 21.55% of 778.663 heads of cattle in East Nusa Tenggara province. This study was aimed at evaluating the potency, constraints and opportunities of beef cattle to small farmers. Two hundreds thirty two respondents were interviewed, site observation was made on the cattle farming. The results showed that the average number of cattle ownership was 9 AU/ farmer. The majority (94.83%) of the farmers on productive age (44 years old) with low education and 17 years farming experience. The main objectives of raising cattle was for breeding (68.53%), fattening (14.22%) and the combination. Animals were commonly kept off shelter, 60.25% of farmers grazed during the day and tethered at night. Natural matings was common, cattle were fed with forages comprises grass and legumes. Additional feed and mineral were given as necessarily. Reproductive characteristics was considerably high, indicated by the ratio of males and females of 1:2.57, high birth rate of 79.69%, and highly adapted of animal to the feed scarcity and harsh environment. The main constraints were the unavailability of feed and sufficient water during dry seasons, high mortality rate of young animal 19.34% due to diseases caused by malnutrition, lack of animal health and disease control as well knowledge and skill of village farmers. Bali cattle has potential to be developed in South Central Timor through the provision of sufficient feed with balance nutrients during dry season, health and disease control by livestock services, and training the farmers on cattle management*

**Keywords:** *cattle, production, small farmers, South Central Timor*

## Introduction

South Central Timor (TTS) is aregencyin the province of East Nusa Tenggara (NTT). Geographically it was located in the southern part of Timor Island with elevation varying from the lowest 44 meters above sea level in Boking districttothe highest1, 480 meters above sea level in Fatumnasi district. TTS regency is characterized as a dry tropical climate with only two seasons, dry and rainy, with an average rainfall in 2012 of 1,054.81 mm/year (BPS Kabupaten TTS, 2014).

Raising beef cattle is part of farmers activity in TTS, a tradition that must be preserved from generation to generation (Ratnawaty and Budianto, 2011). Yusuf and Nulik (2008) reported that beef cattle in this region had a number of roles, namely as: (1) a source of family income, (2) labor replacement, (3) a symbol of social status of family members, (4) a source of fertilizer for plants, and (5) a source of protein for the community. Based on the agriculture census, in 2011 TTS was a center for beef cattle development, with a population of 167.834 heads(21.55%) of the cattle population in NTT province (BPS, 2011). Beef cattle contributed up to19.8% to the total GDP of TTS which reached 2.88 trillion rupiah (approximately 215.5 million USD) in the year 2012. This indicates that TTS has good prospect for beef cattle development. The study aimed to evaluate the potency, constraints and opportunities for improving beef cattle production, for small holding farmers well being.

## Results and Discussion

### Characteristics of Farmers

Men were dominantly responsible for beef cattle farming activity compared to women (94.83%: 5.17%). According to Amalo *et al.* (2012), effective time spent daily for cattle raising activities was 5 hr/d, 6 d a week, making more females preferred to act as supporting labor in such family business. The farmers were categorized in their productive ages averaging 44 years, and farming activities had been introduced to

their children since younger ages 20 year old. A few farmers aged 72 years old still raised cattle, reflecting that beef cattle farming had been carried out from generation to generation. Farmers had averagely 17 years experience in cattle farming. Most of them had low level of formal education background, approximately 61.64% were elementary garduates. BPS data for TTS District showed that TTS community spent only 6.6-6.7 years in formal education during the period 2011-2013 (BPS Kabupaten TTS, 2014). According to Soeprana(2005) and Siswoyo et al. (2013), low education level of farmers could affect their access to information and appropriate management and technology in cattle husbandry, feed and reproduction, and other production input related to institutional roles and development.

Although activities in beef cattle production could improve economic status of farmers, their involvement infarmers groups/institutions was low (39.22%). Despite its importance to strengthening farmers capacity, and bargaining positions. Siswoyo *et al.* (2013), stated that farmers groups was essential in increasing cash income of group members, hence, family welfare.

Table 1. Characteristics of beef cattle farmers in South Central Timor Regency

No.	Variables	Results
1.	Involment cattle farming activity (%)	
	a. Men	94.83
	b. Women	5.17
2.	Age (years)	43.96±11.1
3	farmers' education (%)	
	Elemantary School	61.64
	Yunior High Shool	19.83
	Senior High Shool	15.95
	University	0.43
3.	Farming experiences (years)	17.38±11.0
4.	Involved in farmer groups (%)	39.22
5.	Family size (person/family)	3.66±1.7
6.	Cattle ownership (heads/farmer)	4.94 ± 2.6

### Beef Cattle Production and Management

Most TTS farmers (98.71%) raised Bali cattle as the main commodity in agricultural sectors. Based on BPS (2011), Bali breed was predominat cattle found in NTT, 87.84%. Data in Table 1 showed that 60.53% of farmer owned cattle on average of 4.94 ± 2.6 head/farmer, ranging from 1-10 heads/farmer. Few farmers (3.51%), owned more than 50 heads of cattle with an average of 78.00 ± 25.2 heads/farmer. This figure was doubly higher than the findingof Lake *et al.* (2010), that farmers in Belu District owned an average of 37 cattle/farmer, indicating that beef cattle was the main sources of farmer's income.

As shown in Table 2, Bali cattle were raised for different purposes; those were breeding (68.53%), fattening (14.22%) and combinationof breeding and fattening (17.24%). Rearing systems consisted of tethered all day (18.11%), grazing at daytime and tethered at night (41.81%), and grazing all day (9.48%). Cattle were tethered near the house or in the garden.

Bali cattle was basically fed on farages. The cattle forages compraised native grass (30.98%), cultivated grass (29.38%), legume (25.51 %), and agricultural waste (14.12%). Generally, the forages were obtained from close pastures (35.45%), farmer garden (33.33%), and non-irrigated paddy fields (19.09%). The cultivated grass consisted of mainly *Pennisetum purpurhoides*, *Pennisetum purpureum*, and *Panicum maximum*, whilecommon legumescattle feedwere*Sesbania grandiflora*, *Leucaena leucocephala*, and *Acacia leucophloea*. The agricultural wastes widely used were banana stem, corn straw, green bean straw, cassava straw, and sweet potato straw. Adult cattle, young cattle, and calves were given forage 22.27 kg, 15.65 kg, and 8.25 kg, respectively. Rahmansyah *et al.*, (2013) stated that the concept of cattle rearings utilizing various species plants on dry land obtained fromforest, gardens and dry fields was quite feasible for small-scale farmersin TTS District.

Table 2. Beef cattle production system in South Central Timor Regency (TTS)

No.	Variables (n=228)	Results (%)
1.	Main objective of raising cattle	
	Breeding	68.53
	Fattening	14.23
	Combination breeding and fattening	17.24
2.	Rearing system	
	Tethered all day	18.11
	Grazing day time and tethered at night	41.81
	Grazing all day	9.48
3.	Types of feed	
	Forages	65.04
	Forage and additional feed	28.93
	Additional feed additive	6.03
4.	Matting system	
	Natural matting	70.26
	Artificial insemination	1.72
	Others	28.02
5.	Birth and mortality rate	
	Birth rate	79.69
	Mortality rate of young animal	12.16
	Mortality rate of adult animal	7.18

Natural matings was applied by 70.26% farmers without considering the quality of breeding males, and 28.02% practiced mating systems that related to the raising patterns such as all-day-grazing or day-time grazing. Artificial insemination (AI) method was applied only by few farmers who implemented tethered and stall raising systems near farmers house or in the garden. Reproductive performances such as birth rate (79.69%), mortality rate of calf under one year old (12.16%), and the mortality of adult cattle (7.18%) occurred under such extensive rearing systems. Wirdahayati (2010) reported the average birth and death rates of Bali cattle under one year was 75 and 30% respectively. High adaptability of Bali cattle to the lack of feed and poor environment was one of the advantages to keeping this species in Timor island.

### Constraints and Opportunities

This study showed that the main problems faced by farmers in South Central Timor Regency were the unavailability of feed and drinking water during such a long-critical dry season, poor animal health, and lack of farmers knowledge on breeding and reproductive management. The critical long dry period with during eight months resulted in poor and low quality feed available for animals. According to BPS Kabupaten TTS (2014), within the period of July and November 2013, was the lowest rainfall rate (0-362 mm/year). As a result, mortality rate of calves was (12.16% and adult animal 7.18% (moderate). The other inhibiting factors to cattle development programs included long chain marketing systems, poor feeding and breeding management and limited financial support for breeding program.

Improvements were required for cattle development in TTS, which included capacity building of farmers, provision technology for feeds and feeding, breeding and reproduction, disease control, and strengthening farmers groups to help an easy access to production input and marketing.

### Conclusion

Bali cattle in Central Timor Regency has been well adapted to the local environment, had a relatively good reproduction rate, despite the high mortality rate, was potential to be improved as a source of income for small farmers in the region. Providing good quality feed and fodder through application of simple preservation, appropriate breeding systems and regular health and diseases control by Livestock Service Agency were some alternative solutions needed in improving cattle productivity. Training of farmers on the respective aspects, particularly related to appropriate management of cattle, including strengthening the existing farmers groups or institutions would be required.

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# The Performance of Peranakan Ongole (PO) cattle and Their Crossbreeds in Growing and Fattening Periods

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## Abstract

The local crossbred cattle mainly raised by farmer in Java have a good prospect as a beef producer. This study aimed to compare the performance of PO cattle and their crossbreeds from artificial insemination program using semen of taurus breeds in growing and fattening periods. The first study involved 24 heads of male calves consisting PO (Peranakan Ongole), Simpo (Simmental x PO), Limpo (Limousin x PO) and Simbrah (Simmental x Brahman), and the second study involved 20 heads of male feeder cattle consisting of PO, Simpo, Limpo and Braspo (Brangus x PO). The calves were raised for a ten month period. During pre-weaning period, the calves were raised with their cows and fed 4 kg concentrate and 25 kg King grass per head per day. The calves were weaned at six month old and given approximately 3 kg concentrate and 25 King grass per head per day in a coloni pen. In cattle fattening study, the feeder cattle were kept in individual cage and given 6 kg concentrate ration and ad libitum straw silage for four months. Variables observed included monthly liveweight of calves for a ten month period, pre- and post-weaning growths, feedlot gain, concentrate feed consumption and conversion, slaughter and carcass weights, carcass dressing percentage, and backfat thickness. The result showed that in growing period, the local cross bred cattle grew significantly ( $P < 0.05$ ) faster and than PO cattle, while among the local crossbred cattle, Limpo cattle showed the highest pre-weaning growth and Simbrah cattle the highest post-weaning growth. Compared to PO cattle, the local crossbred cattle had significantly ( $P < 0.05$ ) better feedlot performance in term of concentrate dry matter consumption, daily gain, concentrate feed conversion and final bodyweight. The Braspo cattle produced the highest carcass weight and percentage and the lowest twelfth rib fat thickness relatif to Simpo, Limpo and PO cattle.

**Keywords:** carcass traits, feedlot performances, growth, local crossbred cattle

## Introduction

Increase in domestic demand for beef has led to high importation of this commodity. It was reported that in the year 2012, the total importation of cattle and beef was amounted to 121,616.5 metric ton and valued at 392.9 million dollar (Ditjen PKH 2013). This increased demand of beef could be triggered by increases in population, per capita income, human nutrition status and incoming foreigners (Khasrad dan Ningrat, 2010). Local cattle are still the major source of domestic beef consumption, and yet their productivity are relatively low. Several breeds of local cattle such as Peranakan Ongole (PO), Madura and Bali cattle have been crossed with taurus breeds through artificial insemination programme in order to improve their productivity (Kutsiyah *et al.*, 2003; Hartatik *et al.*, 2009; Depison, 2010). The previous study revealed that the local cattle slaughtered in public slaughter house were mainly in poor (34.7%) and moderat (50.2%) conditions, and only 15.1% in fat condition (Fakultas Peternakan IPB, 2012). This becomes one of the major causes of decrease in domestic beef production since the farmers rarely exercise cattle fattening. In this study, the performance of PO cattle and their crossbreeds were compared in growing and fattening periods.

## Materials and Methods

### *Cattle and Procedures*

Twenty four heads of male calves consisting PO (Peranakan Ongole), Simpo (Simmental x PO), Limpo (Limousin x PO) and Simbrah (Simmental x Brahman) were used to investigate their performance in growing period, and while 20 heads of male feeder cattle aging I<sub>1</sub> dentition, which comprised PO, Simpo, Limpo and Braspo (Brangus x PO) were used to study their performance in fattening period. The calves were raised for a ten month period. During pre-weaning period, the calves were raised with their cows and fed 4 kg

concentrate and 25 kg King grass per head per day. The calves were weaned at six month old and given approximately 3 kg concentrate and 25 King grass per head per day in a coloni pen. In fattening cattle study, the feeder cattle were kept in individual cage and given 6 kg concentrate ration and *ad libitum* straw silage per head per day for four months. One cattle (Braspo) was excluded from the trial due to stomach disorder. The cattle were slaughtered in public slaughter house. They were deprived of food but not water for 24 hours before slaughtering.

### Measurements

The variables observed for cattle during growing period included liveweight on monthly basis for a ten month period, pre- and post-weaning growths. In cattle fattening study, the variables observed included daily gain, concentrate feed consumption and conversion, slaughter and carcass weights, carcass dressing percentage, and back fat thickness on the twelfth rib positio from,  $\frac{3}{4}$  from medial to lateral edge.

### Statistical Analysis

The effect of breed on growth pattern for the first ten months was analysed by repeated measures analysis of variance. The effect of breed on feedlot performance was analysed by analysis of covariance with initial liveweight and time on feed as covariable to examine the breed effect on carcass characteristics (Kaps and Lamberson, 2004).

## Results and Discussion

The growth performance of PO cattle and its crosses from birth to ten month age is presented in Table 1. Significant between breed differences in liveweight occurred in one month age of cattle and thereafter. In general, Simpo, Simbrah and Limpo cattle had significantly ( $P<0.05$ ) higher liveweight than PO cattle and the differences were wider with increasing age. The higher liveweight of the crossbred cattle were due to their superior pre-weaning growth relative to PO cattle, meanwhile only Simbrah showed superior post-weaning growth relative to the other three breed.

Table 1. The effect of local cattle breed on their performance in growing period

Age (month)	Liveweight (kg)			
	PO	Limpo	Simpo	Simbrah
Birth	25	30	32	31
1	38 <sup>a</sup>	53 <sup>ab</sup>	56 <sup>b</sup>	66 <sup>b</sup>
2	49 <sup>a</sup>	78 <sup>b</sup>	78 <sup>b</sup>	86 <sup>b</sup>
3	62 <sup>a</sup>	103 <sup>bc</sup>	94 <sup>b</sup>	111 <sup>c</sup>
4	75 <sup>a</sup>	133 <sup>bc</sup>	120 <sup>b</sup>	137 <sup>c</sup>
5	88 <sup>a</sup>	173 <sup>c</sup>	145 <sup>b</sup>	162 <sup>c</sup>
6	100 <sup>a</sup>	203 <sup>c</sup>	170 <sup>b</sup>	186 <sup>c</sup>
7	114 <sup>a</sup>	216 <sup>c</sup>	187 <sup>b</sup>	206 <sup>c</sup>
8	128 <sup>a</sup>	227 <sup>c</sup>	200 <sup>b</sup>	227 <sup>c</sup>
9	140 <sup>a</sup>	238 <sup>c</sup>	217 <sup>b</sup>	247 <sup>c</sup>
10	157 <sup>a</sup>	248 <sup>bc</sup>	232 <sup>b</sup>	259 <sup>c</sup>
Growth				
Pre-waning (kg/day)	0.42	0.95	0.75	0.87
Post-weaning (kg/day)	0.48	0.38	0.52	0.61
Overall (kg/day)	0.44	0.73	0.67	0.76

SE of liveweight 12.42 kg; Values in the same row followed by a different letter differ significantly ( $P<0.05$ ); PO Peranakan Ongole, Limpo LimousinxPO, Simpo SimmentalxPO, Simbrah SimmentalxBrahman

Brandt *et al.* (2014) reported superiority in the growth rate of the F1 crossbred cattle due to heterosis effect. The higher pre-weaning growth of the crossbred cattle relative to PO cattle indicated better mothering ability of the crossbred cattle in term of milk produced for their calves. Variation in post-weaning growth among the crossbred cattle, particularly lower post-weaning growth of Limpo cattle might be caused by feeding management factor that could not fulfil the requirement of Limpo Cattle for growth.

Table 2. The effect of local cattle breed on their performance and carcass characteristics in fattening period

Parameters	Breed			
	PO	Limpo	Simpso	Braspo
Concentrate Consumption* (kg/head/day)	5.63 <sup>b</sup> ±0.14	4.78 <sup>a</sup> ±0.09	4.78 <sup>a</sup> ±0.07	4.86 <sup>a</sup> ±0.09
Concentrate Feed Conversion	7.80 <sup>c</sup> ±0.38	4.47 <sup>b</sup> ±0.26	4.48 <sup>b</sup> ±0.20	3.73 <sup>a</sup> ±0.26
Daily Gain (kg/head/day)	0.67 <sup>c</sup> ±0.05	1.14 <sup>b</sup> ±0.03	1.13 <sup>b</sup> ±0.03	1.34 <sup>a</sup> ±0.03
Slaughter Weight (kg)	486.90 <sup>c</sup> ±6.94	540.69 <sup>b</sup> ±4.77	539.36 <sup>b</sup> ±3.69	565.77 <sup>a</sup> ±4.73
Carcass Weight (kg)	243.80 <sup>b</sup> ±14.54	281.40 <sup>b</sup> ±13.90	284.60 <sup>b</sup> ±7.40	303.75 <sup>a</sup> ±32.83
Dressing Percentage (%)	48.78 <sup>c</sup> ±1.74	53.72 <sup>b</sup> ±1.29	53.96 <sup>b</sup> ±1.05	56.95 <sup>a</sup> ±1.82
12 <sup>th</sup> Rib Fat Thickness (mm)	0.60 <sup>c</sup> ±0.10	1.42 <sup>b</sup> ±0.08	1.48 <sup>b</sup> ±0.13	1.75 <sup>a</sup> ±0.12

Parameters on feedlot performance corrected for initial liveweight 399.3 kg and time on feed 122.7 days; \*measured in dry matter basis; Values in the same row followed by a different letter differ significantly ( $P < 0.05$ ); PO Peranakan Ongole, Limpo LimousinxPO, Simpson SimmentalxPO, Braspo BrangusxPO

The performance of feedlot cattle at fattening period is presented in Table 2. Compared to PO cattle, the local crossbred cattle consumed significantly ( $p < 0.05$ ) less concentrate, produced significantly ( $p < 0.05$ ) higher daily gain and significantly ( $p < 0.05$ ) more efficient in concentrate feed utilization. Consequently, the crossbred cattle had significantly ( $p < 0.05$ ) heavier slaughter weight, carcass dressing percentage and twelfth rib fat thickness. The superiority of the local cross bred cattle in feedlot fattening had been reported. Soeharsono *et al.* (2011) found daily gain and slaughter weight of 1.28 – 1.63 kg/head/day and 543 – 712 kg respectively for local crossbred cattle in feedlot fattening. The local cross breeds could showed better feedlot performance and more profitable relative to Brahman Cross cattle (Soeharsono *et al.*, 2010). Priyanto *et al.* (1999) reported that increase in slaughter weight of cattle would resulted in increased carcass weight and fatness and therefore saleable beef yield. In addition, it was shown that the cross breed BrahmanxHereford cattle was superior in saleable beef yield compared to the pure breed Brahman or Hereford cattle. The local cattle slaughtered in public slaughter house were mainly in poor (34.7%) and moderat (50.2%) conditions, and only 15.1% in fat condition (Fakultas Peternakan IPB, 2012). Therefore, there is a potency to obviously increase domestic beef production through feedlot fattening of local crossbred cattle.

## Conclusion

The local crossbred cattle showed superiority over the local cattle in growth performance at both growing and fattening periods. This resulted in heavier slughter weight and higher carcass dressing percentage of the local crossbred cattle.

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# Production Performance and Egg Quality of Laying Hens on Cage System with Different Housing Temperature

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## Abstract

High ambient temperature of laying hens in cage system causes high stress and low productivity. Control of ambient temperature becomes an important effort to minimize stress. This research was subjected to evaluate the response of laying hens in cage system to different housing temperatures on production performance and egg quality. A number of 36 laying hens of 30 weeks were observed. The laying hens were placed at 2 different pens in a small closed house at different housing temperatures of 18°C and 30°C. This research was completely randomized designed. Production performance was subjected to t-test, whereas egg quality was descriptively analyzed. The average of hen-day production and feed conversion ratio were not significantly different among treatments. The average of feed consumption, egg weight and egg mass at housing temperature of 18°C were significantly higher ( $P < 0.05$ ) than at 30°C. Income over feed cost was IDR 539.85/hen/day and IDR 393.15/hen/day at 18°C and 30°C, respectively. Haugh Unit, eggshell thickness and eggshell weight at 18°C were higher compared to 30°C. There were no dirty eggs, cracked eggs, and broken eggs at both rearing temperatures. As conclusion, production performance and egg quality of laying hens in cage system at 18°C were better than at 30°C.

Keywords: cage system, egg quality, income over feed cost, laying hens, production performance

## Introduction

Badan Pusat Statistik (2014), stated that the needs of Indonesian people to eggs in 2013 was 1,159,549 tons, and it increased by 5.53% from the previous year. To meet this need, laying hens population in 2013 was amounted to 147.2 million, and it increased by 6.17% from 2012. Therefore the commercial laying hens were one kind of the poultries that very potential in Indonesia.

Environmental factors which have great impact on productivity of laying hens is the temperature of rearing. The comfortable temperature (thermoneutral zone) for laying hens is 20-24°C (Bell and Weaver, 2002). In this temperature range, laying hens will not produce much body heat, so the use of energy becomes more efficient. The temperature change will be responded quickly by laying hens.

The environmental temperature in Indonesia, especially during the daytime (30-34°C), is above the range of comfortable temperatures for laying hens. This is a major constraint in rearing of laying hens. At high temperatures, the chicken release the body heat through panting. Respiratory rate of chicken can increase by up to 200 times/minute (Cunningham and Klein, 2007). The impact of it is a decrease in body resistance, production and quality of eggs produced. Even at extreme temperature ( $\geq 34^\circ\text{C}$ ), can lead to death until 31.7% (Mashaly *et al.*, 2004).

Moreover, the majority of laying hens in Indonesia were reared in cage system (individual cage). This fact make the laying hens more stress. So, the negative impact of rearing of laying hens in the tropics with cage system can be minimized by controlling the ambient temperature of housing. Therefore, the purpose of this study was to find out the response of laying hens toward the difference of rearing temperature in cage system on production performance and quality of eggs produced.

## Materials and Methods

### Animal Experiments and Rearing

The study was conducted in the Poultry Laboratory, Department of Animal Production and Technology, Faculty of Animal Science, Bogor Agricultural University. A number of 36 laying hens from Lohmann strains, 30 weeks age were used. Two small closed house (2 x 2 m<sup>2</sup>) that be equipped temperature controller were used in this study.

Each of pen was filled with 18 chickens that placed in individual cages. Temperature in the first small closed house was set at 18°C, and 30°C in the second of it. Every of it was equipped with light bulb (75 Watt). The feed and water were placed in front of the cage. The feed was commercial feed that contain 14-17% of crude protein, and 2850 kcal/kg of metabolizable energy. The feed was given 120 g/hen/day, but the water was given *ad libitum*.

Rearing was carried out for 6 weeks. Recording of egg production, then they were weighed was done every day. Feed were weighed once a week to calculate feed conversion ratio (FCR). The quality of eggs (Haugh Unit, eggshell thickness, eggshell weight, eggshell wholeness and level of dirty egg) were analyzed in every weekend.

The value of Haugh Unit was calculated by following formula (Keener *et al.*, 2006):

$$HU = 100 \log (H + 7.57 - 1.7 W^{0.37})$$

Information:

HU = value of Haugh Unit

H = high of eggwhite/albumen (mm)

W = egg weight (g/egg)

## Data analysis

Data of production performances were analyzed with t-test using completely randomized design. The temperature of rearing (18°C and 30°C) were as treatments and observation data were as response (Mattjik and Sumertajaya, 2006). Data of egg qualities were descriptively analyzed.

## Results and Discussion

### Performance of Production

Performance of production, including feed consumption, hen day production, egg weight, egg mass, feed conversion ratio and the value of income over feed cost those produced at different temperatures were presented in Table 1.

Table 1. Production performances of laying hens during 6 weeks rearing at different housing temperatures

Performances of production	Housing temperature	
	18 °C	30 °C
Feed consumption (g/hen/week)	830.80 ± 8.80a	818.01 ± 8.46b
Hen day production (%)	89.30 ± 1.50a	87.70 ± 2.90a
Egg weight (g/egg)	62.89 ± 3.27a	60.44 ± 3.19b
Egg mass (kg/6 weeks)	42.46 ± 0.65a	40.07 ± 0.74b
Feed conversion ratio	2.11 ± 0.16a	2.21 ± 0.24a
Income over feed cost (IDR/hen/day)*	539.85	393.15

Note: different letters in the same row indicated statistically significant differences (P < 0.05). \*) Not statistically analyzed

The effect of the different rearing temperatures (18°C and 30°C) in cage system produced the significant difference (P < 0.05) toward the average of feed consumption, egg weight, and egg mass. However, the influence of these temperature difference toward parameters of hen day production and feed conversion ratio were not significantly different.

The average of feed consumption of laying hens that reared at high temperature (30°C) was significantly lower than at 18°C. It was due to the rearing with high temperature, metabolic rate accelerated, so that metabolic heat production increased. Thus, the heat production that must be taken out from the body also increased. These heat production is a signal that be delivered to hypothalamus. This stimulation made the hypothalamus secrete the releasing factor, which subsequently delivered to anterior pituitary. Furthermore, the anterior pituitary produced some hormones, which among others is the gastrin hormone, that have function as mediated negatif feedback. As a result of these mechanism was the occurrence of the decrease of feed consumption (Squires, 2003).

The decrease of feed consumption of laying hens that reared at 30°C resulted in the decrease of the average egg weight, but it had no effect on hen day production. It can be due to genetically the laying

hens have a very high ability to lay eggs. However, the hen day production that produce by laying hens at 18°C and 30°C were not significant statistically, but the egg mass which was obtained during 6 weeks was significantly different ( $P < 0.05$ ).

The value of income over feed cost is the overview of profit. It was a difference between the value of the sale of eggs with the cost of feed. In this study, the laying hens that reared at low temperature (18°C) produced the value of income over feed cost of IDR 539.85/hen/day, and it was in the amount of IDR 146/hen/day higher than the laying hens reared at 30°C.

### Egg Quality

The result of the assay of egg qualities that produced by the laying hens with different temperature of rearing were presented in Table 2.

Table 2. Egg qualities of the laying hens during 6 weeks rearing at different temperatures

Egg qualities	Housing temperature	
	18 °C	30 °C
Haugh Unit	79.20 ± 2.60	74.70 ± 3.80
Eggshell thickness (mm)	0.43 ± 0.04	0.40 ± 0.05
Eggshell weight (%)	12.46 ± 0.65	11.14 ± 0.80
Eggshell wholeness (%)	100.00	100.00
Eggshell dirty (%)	0.00	0.00

Haugh Unit value reflects the level of egg white/albumen thickness. The higher level of egg white thickness, the higher quality of the egg. The laying hens that were reared at 18°C produced the eggs with the higher HU value than those at 30°C. The increase of metabolic rate of laying hens that reared at high temperature caused the process of nutrient absorption was not optimal. Besides that, the increase of metabolic rate also produced higher waste product ( $H_2O$  dan  $CO_2$ ) (Yousef, 1985). It leads to the higher water concentration in eggs (Daghir, 2008). The higher of water concentration in egg caused the lower HU value of eggs that produced by the laying hens at high rearing temperature.

The other impact of the rearing of laying hens at high temperature was the low of eggshell thickness level (Stadelman and Cotterill, 1995). It has been proved in this study. The laying hens that reared at 30°C produced the lower eggshell thickness and the percentage of eggshell weight (0.40 mm dan 11.14%) than those at 18°C (0.43 mm dan 12.46%). Thin eggshell that was produced by the laying hens due to the increase of panting intensity, as an effort to release the heat production from the body. It caused the decrease of  $CO_2$  concentration in the blood. The low of  $CO_2$  concentration in the blood decreased formation process of  $CaCO_3$ , which is a main component of eggshell (Okubo *et al.*, 1997).

In this research, the cracked and broken eggs were not observed at both housing temperatures. All the eggs were categorized into clean egg (based on standard from USDA, 1964). Thereby can be stated that rearing of laying hens in individual cage can minimize the cracked and broken egg.

### Conclusion

Rearing of laying hens in individual cage at 18°C produced higher production performance, egg quality, and income over feed cost value than at 30°C. It was not found the dirty egg, cracked egg, and broken egg on rearing at both temperatures.

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# Evaluation of Good Dairy Farming Practice Implementation In Dairy Goat Farm

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## Abstract

*The present study was conducted to evaluate the implementation of Good Dairy Farming Practice (GDFP) in dairy goat farms in Bogor regency West Java Indonesia. Four farms having more than 100 dairy goats were purposively selected for the study. Aspects of Good Dairy Farming Practices (GDFP) observed were: (1) breed and reproduction, (2) feed and drinking water management, (3) farm management, (4) housing and equipment, (5) animal health, and (6) animal welfare. Score of 4 (very good), 3 (good), 2 (fair), 1 (poor), and 0 (very bad) were used to assess each aspects. This study revealed that Etawa crossbred goats (PE) were more preferable than others dairy goat types such as Saanen, due to its' higher in milk production. Farmers already provide forages mixed with other feed materials with better nutritional content. Farmers do not have a complete business records, and yet have a farm waste processing unit. Farmers were already concerned about the health and welfare of the goats. The average score for breed and reproductive aspect was 3.54; feed and drinking water management aspect was 3.57; farm management aspect was 3.39; housing and equipment aspect was 3.17; animal health aspect was 3.53; and animal welfare aspect was 3.63. Conclusion of this study was the GDFP application in all of dairy farms studied the studied were very good.*

**Keywords:** dairy goat, good dairy farming practice

## Introduction

Goat milk agribusiness development must be supported by the ability of the goat dairy farms to fulfill consumer' demand and expectations. Currently, there are no data or complete documentation about dairy goat Goat milk agribusiness development must be supported by the ability of the goat dairy farms to fulfill consumer' demand and expectations. Currently there are no data or complete documentation about dairy goat population, the number of dairy goat farms, total production and goat milk market share in Indonesia. Market demand for goat milk has increased in the last few years, but has not been met due to the limitation in production. This is due to the low population of existing dairy goat, and farmers do not applying the principle of good farming practices yet as well. This study was conducted to evaluate the implementation of Good Dairy Farming Practice (GDFP) in dairy goat farms in Bogor District. The results of this study are expected to provide information to all parties involved in goat milk agribusiness about the existing problems, and how the solution can be done for dairy goat farms development in the future.

## Methods

The research was conducted in four dairy goat farms in Bogor District, West Java Province. The samples were chosen purposively i.e. farms having more than 100 dairy goats. The aspects of Good Dairy Farming Practices (GDFP) observed were breed and reproduction, feed and drinking water management, livestock management, housing and equipment, animal health, and animal welfare. Assessment of GDFP aspects was measured by giving a score of 4, 3, 2, 1 and 0. The value of every aspect then summed and the average is calculated. Classification of farm performance was generally viewed from the average score, and then grouped as follows:

1. If the value of the average performance of the farm is from 0.00 to 1.00 it means that the application of GDFP in the farm is bad;
2. If the value of the average performance of the farm is from 1.01 to 2.00 it means that the application of GDFP in the farm is fair;
3. If the value of the average performance of the farm is from 2.01 to 3.00 it means the application of GDFP in the farm is good;
4. If the value of the average performance of the farm is from 3.01 to 4.00 it means that the application of GDFP in the farm is very good.

## Results and Discussion

Good Farming Practice (GFP) according to the Ministry of Agriculture (Kementan, 2010) is a guideline that describes how the cultivation of plants and animals well in order to produce food-grade, safe and suitable for consumption. Based on the FAO and IDF (2011) aspects that should exist in the implementation of Good Dairy Farming Practice (GDFP) are animal health, milking hygiene, nutrition/feed and water, animal welfare, environment, and socio-economic management.

### Aspects of breed and reproduction

Table 1 shows that GDFP application score for breed and reproduction aspects in the studied farms were very good (average score was 3.54). The types of dairy goats that most widely kept in studied farms were Etawah grade (Peranakan Etawah/PE). Etawah goat is a type that has high productivity and good durability. PE goats are common in Kaligesing, Purworejo Central Java. PE is the result of grading-up mating between *Kacang* and Etawa goat (Sodiq & Tawfik, 2004). The age of dairy goats in the analyzed farm were between 3-3.5 years (I3-I4), or in the third and fourth lactation. Age and lactation period of goat were determined by estimation of its mandibular incisors. Selection is a very important activity in dairy goat farm. Selection that based on visible observation was very common at the traditional livestock market, both by farmers and traders or appraiser. All studied farms usually select young doe and superior male for breeding stock at buckling and doeling stage. Guidelines for choosing a good dairy goat breed were depending on the purpose and the business interests of each farm. Selection in the studied farms is done by assessing the performance based on several benchmarks including: ear shape, body length, face shape, and udder postures. All farmers already had a good knowledge of when the goat estrus, and when a goat should be mated again after kidding.

### Dairy Goat Feed and Drinking Water Management

Average score for dairy goat feed and drinking water was 3.57 (very good). To maintain optimum milk production and good health, farmers fed their goats with a balanced diet for energy, protein, minerals, and vitamins, according to their needs. Feed nutrients are used for growing, reproduction, lactation, and movement. The composition of forage and concentrates must be balanced, and three kinds of forages such as grass, legumes and pasture should constitute a majority of the daily diet (Basitan & Jarcia, 2013; Fuah & Pattie, 2013; Morand *et al.*, 2007; Sodiq & Setianto, 2009). Farmers usually feed their goats with a variety feeds. Farm A and D provides their goats with Napier grass (*Pennisetum purpureum*), field grass and other forages, and concentrates (i.e. dregs of beer, tofu waste or bran) as well. With the average of 4-5kg/day of forage and approximately 400-500g/day concentrates are given as goat daily intake. Whereas farm B gives concentrate mixed with cassava, bran, pulp, and dry onion skins they purchased from markets around their farms in Bogor. In addition, forages consisting of grass and leaves of cassava, *calliandra*, *gliricidia* and *indigofera* are also given. Finally the farm C feeds its goats with forages and concentrates. Forages given are Napier grass (*Pennisetum purpureum*). In addition, concentrate containing soybean cake waste, tofu waste, palm waste, black cumin (*Habatussauda*) and ready-made concentrate are also given. Only farm D that did not give *ad libitum* drinking water.

### Management

The application for management aspects in the analyzed farms were very good (average score is 3.39). Maintenance of dairy goats should be done as much as possible to maintain the viability and productivity of the livestock. In general, the maintenance of goat adapted to the respective phases of life, ranging from kid, young goat into adulthood. All analyzed farms were holding an intensive system. All farmers were

following full hand method. Farmers were following scientific milking practice and take care of cleanliness as reflected by full hand milking practices and using clean milk utensils. Milking is done twice a day in the morning and afternoon. The average daily milk production is between 1-1.5 liters/head. This study revealed that all farms do not have a complete business records, and a waste processing unit yet.

Table 1. Scores of some aspects implementation toward the Good Dairy Farming Practices on several farms at Bogor Regency

No.	GDFP Aspects	Farms				Average Score
		A	B	C	D	
1.	<b>Breeding and Reproductive</b>					
a.	Dairy goat breed	4.00	4.00	1.00	1.00	2.50
b.	Selection system	4.00	4.00	4.00	2.00	3.50
c.	Mating system	3.00	3.00	3.00	3.00	3.00
d.	Estrous knowledge	4.00	4.00	4.00	4.00	4.00
e.	First mate age	4.00	4.00	4.00	4.00	4.00
f.	Re-mating after giving birth	4.00	4.00	4.00	4.00	4.00
g.	Kidding interval	4.00	4.00	4.00	3.00	3.75
	Average score	3.86	3.86	3.43	3.00	3.54
2.	<b>Feed and Drinking Water</b>					
a.	Forages feeding system	4.00	4.00	4.00	4.00	4.00
b.	The amount of forages	4.00	4.00	4.00	3.00	3.75
c.	Forages quality	3.00	3.00	3.00	3.00	3.00
d.	Forages feeding frequency	4.00	4.00	4.00	4.00	4.00
e.	Concentrate feeding system	4.00	4.00	4.00	4.00	4.00
f.	The amount of concentrates	4.00	4.00	4.00	4.00	4.00
g.	Concentrate quality	3.00	4.00	3.00	3.00	3.25
h.	Concentrate feeding frequency	4.00	4.00	4.00	4.00	4.00
i.	Provision of drinking water	2.00	4.00	4.00	1.00	2.75
	Average score	3.43	3.86	3.71	3.29	3.57
3.	<b>Management</b>					
a.	Goat bathing frequency	4.00	4.00	4.00	2.00	3.50
b.	Bathing system	3.00	3.00	2.00	4.00	3.00
c.	Housing cleaning	2.00	2.00	4.00	4.00	3.00
d.	Milking system	4.00	4.00	4.00	3.00	3.75
e.	Milk post-harvest handling	4.00	4.00	4.00	3.00	3.75
f.	Young doe rearing	4.00	4.00	4.00	4.00	4.00
g.	Dry-off doe	4.00	4.00	4.00	3.00	3.75
h.	Farm recording	2.00	3.00	2.00	4.00	2.75
i.	Waste management	3.00	4.00	2.00	2.00	2.75
	Average score	3.29	3.57	3.43	3.29	3.39
4.	<b>Housing and Equipment</b>					
a.	Housing layout	4.00	4.00	3.00	2.00	3.25
b.	Housing construction	4.00	4.00	3.00	1.00	3.00
c.	Drainage system & sewer	3.00	4.00	3.00	2.00	3.00
d.	Dung shelter	4.00	4.00	3.00	2.00	3.25
e.	Farm equipment	4.00	4.00	4.00	2.00	3.50
f.	Milking equipment	2.00	3.00	4.00	3.00	3.00
	Average score	3.50	3.83	3.33	2.00	3.17
5.	<b>Animal Health</b>					
a.	Knowledge about diseases	4.00	4.00	4.00	3.00	3.75
b.	Diseases prevention	4.00	4.00	4.00	3.00	3.75
c.	Diseases treatment	4.00	4.00	4.00	4.00	4.00
	Average score	3.58	3.81	3.89	2.83	3.53
6.	<b>Animal Welfare</b>					
a.	Freedom from thirst, hunger & malnutrition	4.00	4.00	4.00	4.00	4.00
b.	Freedom from discomfort	4.00	4.00	4.00	3.00	3.75
c.	Freedom from pain, injury and disease	4.00	4.00	3.00	3.00	3.50
d.	Freedom to express normal patterns of behavior	4.00	4.00	4.00	3.00	3.75
e.	Freedom from fear and distress	4.00	4.00	3.00	2.00	3.25
	Average score	3.93	3.97	3.65	2.97	3.63

## Dairy goat housing and equipment

Table 1 shows that GDFP application score for housing and equipment aspects in the studied farms were very good (average score is 3.17). Goat's housing are made in accordance with technical requirements, particularly enable to provide comfort, health and good milk productivity. Farmers use housing materials which is easily available in the surrounding area. Planting trees around the housing at farm A and B make housing more airy, in addition to withstand the wind. Good ventilation, lighting and comfort are factors to be considered to improve the performance of dairy goats (Alcedo *et al.*, 2014; Sabapara *et al.*, 2014). Existing facilities and equipment at the dairy goat farm is milk buckets, funnels, filter, rags, napkins, hoes, sickles and shovels, nail clippers, medical equipment and drug. The equipment and technology should follow standardizes milking procedures to guarantee the good quality of goat milk. Farmers always control all the milking equipment and other facilities regularly. Unclean facilities will be harmful to goats due to the contamination caused by microbial pathogens, chemicals and physical solvents directly and indirect (Gustiani, 2009; Olechnowicz & Sobek, 2008; Taufik *et al.*, 2011). To minimize the occurrence of microbial contamination in milk, the milk packaging room on all farms was located quite close to the milking parlor.

## Animal Health

GDFP application for animal health aspects in the analyzed farms were very good (average score is 3.53). Until now mastitis is still a major problem that cannot be overcome by the farmer, both in dairy cattle and dairy goats. Mastitis has been widely reported to cause losses, especially cause reduced milk production, milk quality damage, and even cause udder gland malfunction (Adriani, 2010; Bourabah, 2013; Koop *et al.*, 2010; Leitner, 2004). Farmers in the analyzed farms have done many attempts to reduce the occurrence of mastitis in their farm. They clean the pens routinely, and cleaning the animal body before milking. In addition to mastitis, diseases that often affect dairy goats are respiratory disorders and parasitic infections. Parasite control is an important consideration in the welfare of all livestock and appropriate action should be undertaken to control and/or prevent parasitic infection. Farmers always consult with a veterinarian to maintain the health of their livestock.

## Animal Welfare

Average score for dairy goat feed and drinking water was 3.63 (very good). According to Broom (2011) and FAWAC (2003) all farms must have proper animal handling facilities including pens and a crush where an animal can be restrained with minimum risk of injury or stress. Farmers have tried to make the animal feel comfortable and safe. Pens were constructed using materials that will not harm livestock. Farmers still trying to provide space so that the goats can freely move, despite of limited land.

## Conclusion

In general the GDFP application in several dairy goat farms at Bogor Regency was very good. Good dairy farming practice application in the studied farms ensures that milk is produced by healthy animals in sustainable and responsible manners with attention to the animal welfare, social, economic and environmental perspectives. So implementation of good dairy farming practice will reduce the management risk for the short and long term future of the dairy farming enterprise.

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# Performance of Chicken Broiler Using Water Hyacinth as a Substitute for Some Rations

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## Abstract

The purpose of this study was to determine the performance of chicken broiler using water hyacinth as a substitute for some rations. This study used 80 broiler chickens strain MB-202P from PT Multi Breeder Indonesia Tbk. aged 2 weeks and were randomly allocated into five treatment groups: R0 = control (without water hyacinth), R1 = 4% water hyacinth, R2 = 8% water hyacinth, R3 = 12% water hyacinth, R4 = 16% water hyacinth, respectively. Each treatment was consisted of four replicates with four chickens per unit. Chickens broiler were given feed and water ad libitum until 6 weeks old, and the remaining feed was weighed every day. The results showed that the performance of broilers using water hyacinth as a substitute for some of the ration had very significant difference ( $P < 0.01$ ) to ration consumption, body weight gain increased, efficient use of rations, carcass percentage, abdominal fat percentage, liver weight, gizzard weight, length of intestine, and total cholesterol. From the results, replacement of water hyacinth on partial rations of 4% - 16% was significantly decreased ration consumption, body weight gain, efficiency of use of the ration, the percentage of carcass, abdominal fat, liver weight, total cholesterol broilers, but higher levels of water hyacinth was found to increase broiler gizzard weights.

**Keywords:** chicken meat. water hyacinth.

## Introduction

Poultry feed is generally derived from agricultural products or byproducts of agricultural products. In a further development of the building blocks of feed is still much to compete with human needs so that in the preparation of food, both quality and quantity consequently cost very high.

Feed costs contribute to the cost of production about 70% (Zuprizal and Kamal, 2000). Therefore it is necessary for efforts to obtain alternative feed ingredients that have not been used but contains nutrients that are not much different from that normally used for feed ingredients.

One way to obtain alternative feed ingredients is the use of wild plants that live in watery area. From one side of the plant is cause more problems both ecologically and economically, this plant can be disturbed if not controlled by irrigation, water disrupting traffic, polluting resources and fishing areas that can lead to flooding and salting of the lake. One of the plants that are included in the group is the water hyacinth plant (*Eichornia crassipes*).

Water hyacinth can be used as animal feed because it contains nutrients which is quite good. Laboratory analysis results from Feed Science and Technology, Faculty of Animal Science, IPB (2011), show that water hyacinth contains dry matter = 90.77; ash = 15.80; crude protein = 8.25; crude fiber = 29.67; crude fat 0.89; extract without nitrogen = 36.13; Ca = 1.18; P = 0.54; NDF = 68.30; ADF = 54.32; hemicellulose = 13.98; cellulose = 46.12; silica = 7.87; lignin; 0.32 energy content (gross energy) hyacinth 3218 kcal.

## Materials and Methods

The research was conducted through an experiment carried out at the site of the Faculty of Animal Research, University of Sam Ratulangi. The experiment was carried out from early September to October 2012. This study used 100 chickens broiler strain MB-202P from PT Multi Breeder Indonesia Tbk. aged 2 weeks.

## Foodstuffs

Material constituent eat rations consisting of yellow corn, coconut cake, fine bran, soybean meal, fish meal, blood meal, bone meal and Hyacinth. Ration was arranged so that the same nutrient content for each treatment. Ration composition experiment, the content of nutrients and metabolizable energy in the ration of each treatment are listed in Table 1, 2, and 3. Water hyacinth that used in this research came from the lake Tondano. Water hyacinth was dried and powdered as flour.

This study used 25 units of battery cage system with dimension 30 x 90 x 30 cm. Each unit was occupied by 4 hens and equipped with feed and drinking facilities. Other equipment used are buckets, knives, plastic bags and scales. This research was conducted through experiments using completely randomized design (CRD) (Steel and Torie, 1993), as well as the treatment consisted of 5 treatments and 4 replications. Placement of chicken into each unit enclosure was done randomly. As the treatment in this trial was the provision of water hyacinth in the ration of broiler with the following structure:

R0 = (without hyacinth)

R1 = 4% hyacinth

R2 = 8% hyacinth

R3 = 12% water hyacinth

R4 = 16% water hyacinth

Table 1. Nutrients Composition Experiment of Food Rations

Nutrients and ME	Treatments				
	R0	R1	R2	R3	R4
Protein	22.93	22.35	21.76	21.17	20.58
Fat	6.48	6.26	6.03	5.81	5.59
Crude Fibre	4.43	5.44	6.45	7.46	8.47
Calcium (Ca)	1.012	1.019	1.025	1.032	1.039
Phosphor (P)	0.872	0.858	0.845	0.832	0.818
ME (k.kal/Kg)	3063.6	3069.7	3075.9	3082.1	3088.3

The variables used to determine the performance of broiler chickens in this study were:

1. Consumption ration (grams), which was obtained from the difference between the amount of rations given to the rest of the ration every day.
2. Carcass, obtained from carcasses weighing without feathers, blood, head, legs, abdominal fat, and offal.
3. Abdominal fat, abdominal fat was measured by weighing, while the percentage of abdominal fat obtained from the ratio between the weight of abdominal fat to body weight and then multiplied by 100%.
4. The weight of the liver
5. The weight of gizzard
6. Length of Intestine

## Result and Discussion

Research data broiler performance which includes feed consumption, body weight gain, feed efficiency, carcass percentage, abdominal fat, liver weight, the weight of gizzard, intestine length and total cholesterol were presented in Table 2.

Table 2. Treatments of Variable Mean Observed During Research

Variables	Treatments				
	R0	R1	R2	R3	R4
Ration consumption (g)	84.16 <sup>a</sup>	80.85 <sup>b</sup>	76.94 <sup>c</sup>	72.01 <sup>d</sup>	67.60 <sup>e</sup>
Carcass percentage (%)	73.62 <sup>ab</sup>	71.30 <sup>bc</sup>	68.46 <sup>c</sup>	58.46 <sup>cd</sup>	56.37 <sup>e</sup>
Abdominal Fat (%)	3.44 <sup>a</sup>	2.73 <sup>b</sup>	2.50 <sup>c</sup>	1.45 <sup>d</sup>	1.19 <sup>e</sup>
Liver weight (g)	26.22 <sup>a</sup>	24.32 <sup>b</sup>	21.29 <sup>c</sup>	20.32 <sup>d</sup>	18.26 <sup>a</sup>
Gizzard weight (g)	15.94 <sup>a</sup>	19.55 <sup>b</sup>	22.20 <sup>c</sup>	24.60 <sup>d</sup>	29.57 <sup>e</sup>
Length of intestine (cm)	166 <sup>a</sup>	171 <sup>b</sup>	179 <sup>c</sup>	183 <sup>d</sup>	187 <sup>e</sup>

Remarks: 1. The same Superscript on the same line shows the difference were not significant (P < 0.05); 2. Superscript different on the same line indicate significant differences (P < 0.01)

## Ration Consumption

The average consumption of broiler ration during the study were in the range of 67.60 g / tail 84.16 g / head, wherein the amount of feed intake was obtained in treatment R4 lowest (basal diet + 16% hyacinth) and consumption the highest in treatment R0 (basal diet). Revelation (2004) states, the consumption of broiler finisher recommended 69-104 grams per head per day, for broilers females 55-76 and males grams 77-104 grams per head per day. According to NRC (1994) and the Poultry CRC (2006) reared broilers aged 2-6 weeks rations consumption ranged between 70-163 grams (for males) and 63.43 grams -143 (for females). Analysis of the performance, indicated that the treatment gave a significant influence ( $P < 0.01$ ) on feed consumption. Test comparisons between treatments by HSD showed that the average feed consumption of each treatment was highly significant ( $P < 0.01$ ) between one another, where R0 feed consumption was the highest among other treatments, and then sequentially followed by treatment of R1, R2, R3 and the lowest consumption was R4 (or  $R0 > R1 > R2 > R3 > R4$ ).

Feed consumption was influenced by the quality and palatability of feed (North, 1984). Cherry (1992) stated that the higher crude fiber in the ration of white leghorn cause the amount of feed intake decreases, because the ration was bulki and absorbed water. Alvarado, *et al.* (2008) suggested a fibrous diet could slow the feed rate of proventriculus and gizzard into the small intestine, because it was influenced by the nature bulki (meeting room digestion) of crude fiber, which could lead to low levels of consumption during the study.

## Carcass percentage

Data carcass percentage during the study ranged 56.38% - 73.62%. The results obtained from this research was still adrift slightly lower than reports McNitt (1983) which stated that the percentage of broiler chicken carcass weight were normal range between 65-75% of the live weight. Analysis of the performance, showed that the use of water hyacinth in the ration provided a very real effect ( $P < 0.01$ ) on carcass percentage. HSD test showed that the percentage of carcasses in treatment R0 significantly different ( $P < 0.05$ ) higher than the treatment of R2, was also highly significant ( $< 0.01$ ) higher than the treatment R3 and R4. But between treatment R0 and R1 treatment with 4% level hyacinth carcass percentage had no significant difference ( $P > 0.05$ ). Similarly, between treatments R1 and R2 there was no significant difference ( $P > 0.05$ ). But the percentage of carcasses of the two treatments (R1 and R2) showed highly significant ( $P < 0.01$ ) higher than the treatment R3 and R4. While between R3 and R4 treatment showed no difference ( $P > 0.05$ ). Jull (1972) said that the carcass percentage was determined by the amount of wasted parts of the body such as the head, neck, feet, viscera, feathers and blood. Siregar (1980) suggested the carcass percentage was the ratio between the carcass weight live weight multiplied by 100%. Mutardjo (1987) explained that the production of carcass closely related to body weight, with increased body weight then followed by an increase in carcass weight. According to Haroen (2003) achievement of carcass weight was closely associated with the slaughter weight and body weight gain. This study showed that the use of water hyacinth as a partial replacement ration at the rate of 8% above the basal ration, suppressed the formation of broiler carcass tissue, so that the percentage of carcasses in the treatment of R2, R3 and R4 showed adrift lower than R0 treatment. Similarly, the treatment of R3 and R4, which carcass percentage had no difference between the two treatments, but different lower compared with the treatment of R1 and R2. It was thought to be caused by an increase in the content of crude fiber in the ration treatment. Correspondingly, Shahin and Abdelazim (2005) stated that the broilers were fed coarse fibrous high (using clover hay) resulted in lower carcass weight compared to broilers fed low crude fiber.

## Abdominal Fat Percentage

Abdominal fat is fat that lies between proventriculus, gizzard, duodenum and around the cloaca. The percentage of abdominal fat obtained based on the distribution of abdominal fat weight to body weight multiplied by 100 percent. Data observation abdominal fat percentage during the study (Table 4) were in the range of 1.19% - 3.44%. Leenstra, *et al.* (1986) suggest that abdominal fat percentage ranged between 2-3% by weight of life. Analysis of the performance, showed that the treatment was highly significant ( $P < 0.01$ ) against the percentage of abdominal fat. HSD test based on a percentage of abdominal fat ration showed treatment R0 highly significant ( $P < 0.01$ ) higher than the R1, R2, R3 and R4. In successive gradual decreased abdominal fat percentage in experimental animals by increasing the proportion of water hyacinth in the diet-ration treatment R1, R2, R3, and R4, were highly significant ( $P < 0.01$ ) between treatments, namely  $R1 < R2 < R3 < R4$ .

The reduced abdominal fat percentage in the treatment ration R1, R2, R3, and R4, thought to be caused by an increase in the percentage content of crude fiber and cellulose, hemicellulose, and lignin silica due to

the increasing standard of water hyacinth on each of these treatments. The results were consistent with the proposed Sutardi (1980), which said that crude fiber absorbs fat in the chicken body so that the effect on the content of abdominal fat percentage was generated. Crude fiber that contained lignin and cellulose stodge (Sutardi, 1980). The cell walls were not soluble in the solvent detergent and divided into several factions based solvent detergent solubility in acid, comprising soluble fraction of the cell wall hemicellulose and protein, whereas insoluble was the acid detergent fiber (Acid Detergent Fiber = ADF), in addition to organic material cell walls also contains silica.

### **Liver Weight**

The range of the liver weight during the study range of 18.26 and 26.22 g. Lowest liver weights contained in R4 (basal diet + 16% hyacinth) and highest in R0 (basal diet). If seen from the proportion of the weight of the liver of live weight broiler in this experiment, the value was between 1.94 to 2.02%. Results were still at the value stated percentage Nickel, *et al.* (1977) and Putnam (1991), which ranged from 1.7 to 2.8% of the live weight.

Analysis of the performance, showed that the change in the percentage of liver weight was very significant ( $P < 0.01$ ) influenced by the treatment. HSD test based on a careful weight ration treatment showed R0 highly significant ( $P < 0.01$ ) higher than the R1, R2, R3 and R4. Furthermore, in all treatments that used water hyacinth R1, R2, R3, and R4 respectively, showed weight loss response liver by increasing the proportion of water hyacinth in the ration. where a decrease in heart weight reaching a highly significant difference ( $P < 0.01$ ) between treatments, namely  $R1 < R2 < R3 < R4$ . Ressang (1984) said that the liver plays a role in the secretion of bile, metabolic fat, iron metabolic, detoxification, formation of red blood cells and storage of vitamins. Further stated that due to the balance of the absorbed nutrients feed the less, the function of the liver was not working optimally to affect the development of liver. After all was said, the liver was the organ components in percentage tends to decline with age. Lesson and Summer (1980) stated that the liver was the organ that grew more rapidly at the age of one week after hatching and the growth declined in adulthood.

### **Gizzard weight**

Gizzard weights were in the range of 15.94 and 29.57 grams. Lowest gizzard weights contained in R0 (basal diet) and highest in R4 (basal diet + 16% water hyacinth). Putnam (1991) stated that the weight of the broiler gizzard ranged from 1.6 to 2.3% of the live weight.

Analysis of the performance, indicated that the use of water hyacinth as a partial replacement ration was highly significant ( $P < 0.01$ ) on the weight of gizzard. HSD test gizzard weights based on ration treatment showed R0 highly significant ( $P < 0.01$ ) lower than the R1, R2, R3 and R4. All animal experiments were received rations treated using water hyacinth R1, R2, R3, and R4 turns increased gizzard weights in a row along with the increasing proportion of water hyacinth in the ration, where the increase reached a highly significant difference ( $P < 0.01$ ) between one treatment to another. namely  $R1 < R2 < R3 < R4$ . The results obtained during this study were consistent with the experiments performed Nasem, *et al.* (2006) on the broiler finisher using canola meal that was high coarse fiber in the ration. which found an increase in weight of gizzard with the increase of crude fiber from the use of canola meal in the ration. Hetland and Svihus (2001) stated that the ration crude fiber components were included in the gizzard undergo a process of grinding up the finer particle size before leaving the gizzard. This condition caused the contents of gizzard volume increased in line with rising crude fiber in the ration. According to Alvarado, *et al.* (2008) rations using the resources of crude fiber properties and water binding capacity (Water Holding Capacity), where the nature of the effect bulky in the gizzard while reducing the rate of proventrikulus and gizzard food into the small intestine, which in turn causes enlargement of the capacity gizzard.

### **Length of Intestine**

The long-range intestine during the study ranged 166 and 175 cm. Intestinal length lowest for the R0 (basal diet) and highest in R4 (basal diet + 16% water hyacinth). The average length of the colon in this study was higher than the percentage of intestinal length Ologhobo research, *et al.* (1993). Analysis of the performance (Appendix 8), showed that the use of water hyacinth as a partial replacement ration gives highly significant effect ( $P < 0.01$ ) to the length of the colon.

HSD test (Tukey test) denoted the length of the intestines in treatment R0 highly significant ( $P < 0.01$ ) shorter than the R1, R2, R3 and R4. Furthermore, in a row there was an increase in the length of the intestines of cattle experiment with increasing the proportion of water hyacinth in the diet-ration treatment

R1, R2, R3, and R4, where the result reached the level of a highly significant difference ( $P < 0.01$ ) between treatments, namely  $R1 < R2 < R3 < R4$ .

Results of this study showed higher concentrations of water hyacinth than the longer ration chicken intestine. The conditions reinforce the notion that the crude fiber content of water hyacinth in the diet affected the small intestine digestive organs to relax in order to allow the absorption of nutrient substances that accumulate with coarse fiber components, causing a lengthening in this digestive organ. Khempaka, *et al.* (2009) reported that the increase in crude fiber rations of dried cassava skin had no lead to an extension of duodenum part of the small intestine. but precisely obviously led to an extension in the jejunum and ileum of the small intestine of broilers, compared with the control diet. Correspondingly, Mourao, *et al.* (2008) reported that the relative length of the small intestine of broilers were given rations of fibrous using citrus peel powder (10%) turned out to be significantly different longer than broilers are using feed controls. According to Ressang (1984) that an increase in the intensity of intestinal peristalsis would increase the length of the intestine. While, Amrullah (2004) argued, the greater the change in bowel and length, followed by the increase in the number of intestinal villi and the secretion of digestive enzymes-enzymes affected overall crude fiber.

## Conclusion

Replacement of water hyacinth on partial rations of 4% - 16% was decreased ration consumption, body weight gain, ration efficiency, carcass percentage, abdominal fat, liver weight, total cholesterol broilers, but higher levels of water hyacinth was found to increase gizzardweights.

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# Chemical and Physical Properties of Rex and Satin Rabbits Meat

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## Abstract

Rabbit meat is known as a healthy meat because it contains high protein with low fat, energy and cholesterol. Today, consumers are likely to eat meat with low fat. Rabbit meat will be able to meet the needs of the market. This study aimed to evaluate the chemical and physical properties of Rex and Satin rabbits meat. Completely randomized design with breed of rabbits by six replications were used to analyzed chemical properties of rabbit meat, and factorial design (2x2) with breed (Rex and Satin) and muscle disposition (*Biceps femoris* and *Longissimus dorsi et lumbarum*) by five replication were used to analyzed physical properties. The chemical properties of rabbit meat were not affected by breed of rabbits (moisture, crude protein, crude fat, Energy, coarse fiber, Ash, Calcium, Phosphorus and total cholesterol). There were no differences on pH and cooking loss between breed and type of muscle, but water holding capacity of *Longissimus dorsi et lumbarum* of Rex rabbit ( $67.52 \pm 14.73\%$ ) was lowest and tenderness of *Longissimus dorsi et lumbarum* of Satin rabbit ( $1.11 \pm 0.28$ ) was most tender.

**Keywords:** chemical properties, physical, rabbit meat

## Introduction

Rabbit is one commodity that is a good producer of meat to be developed because the rabbit is easy to breed and grow relatively quickly so that the rabbit is possible to meet the shortage of meat supply. Besides that rabbit meat has a low cholesterol content, protein content better and rabbit meat is also healthier when compared to other meat of livestock. Gillespie (2004) explained that rabbits have a better quality of meat is high protein content (20.1%), fat content (2.5%) and cholesterol (1:39 mg / kg) is low. Chan et al. (1995) compared the nutrient content of some livestock, that rabbit meat has a protein content (21.9 g / 100g) high, cholesterol (53 mg / 100g) and calories (136 kcal) is the lowest among chicken, beef, sheep and pig. Similarly, with Nistor *et al.* (2013) shows, rabbit meat was richer in calcium (21.4 mg / 100 g) and phosphorus (347 mg / 100 g) than chicken, beef and pork meat, and lower in fat (9.2 g / 100 g) and cholesterol (56.4 mg / 100 g).

Rabbit farmer usually raised popular type in community like Rex and Satin, that people kept them as pet animal. Rex rabbits are known as the pet with quality hair softly as velvet, has a weight of 2.7-3.6 kg on adult age, Satin rabbits are known have shiny hair, smooth, thick and dense and has a different hair length between down hair and guard hair, male of Satin could reached 3.9 -4.8 kg adult body weight and the female could reached 4.1-5.0 kg adult body weight. Both of these rabbits can be used to take skin hair (fur) and potential as a producer of meat (Brahmantiyo *et al.* 2010).

Chemical and physical properties of Rex and Satin rabbits meat is still limited. The purpose of this study was to evaluate the physical and chemical characteristics Rex and Satin rabbits meat.

## Materials and Methods

A total of 12 male rabbits, half from the Rex and half from Satin rabbits, were used in this study. Rex and Satin Rabbits were raised at Indonesian Research Institute for Animal Production. The animal were randomly assigned to a completely random design with six replication for chemical properties analyzed on longissimus dorsi et lumbarum muscle. Complete randomly design with a 2 x 2 factorial arrangement of treatments (two breeds and two location of muscles there were longissimus dorsi et lumbarum and biceps femoris muscles) with five replication for physical properties analyzed.

Animal were slaughtered by halal methods at 24 weeks of age after 12 hours fast. Rabbits were slaughtered at the same time and followed by bleeding, skinning, and cutting on commercial part (Blasco and Ouhayoun, 1996). Longissimus dorsi et lumbarum muscles were dissected for chemical properties

analyze and longissimus dorsi et lumbarum with biceps femoris muscles were dissected for physical properties analyze.

After 24 hours aging period, the muscles were analyzed at Analytical Laboratory at Indonesian Research Institute for Animal Production for chemical properties analyze, and Large Ruminant Laboratory, Faculty of Animal Science, Bogor Agricultural University for physical properties analyze. Chemical properties, such as water, crude protein, crude fat, and energy were measured according to AOAC (1999). pH value of rabbit meat was measured using pH meter (Hanna HI99163), water holding capacity (WHC) was measured according to Hamm (1960). Warner Blatzler Shear Force (WBSF) device was used to measure rabbit meat tenderness. Rabbit meat samples from loin part were used to analyze the cooking loss. The sample was weighed and cooked until internal temperature of 81 °C. Cooked samples were allowed to cool in room temperature for 24 hours. The samples were weighed and recorded into the formula as cooking loss value (%).

$$\text{Cooking loss (\%)} = \frac{\text{raw sample weight (g)} - \text{cooked sample weight (g)}}{\text{raw sample weight (g)}} \times 100 \%$$

Anova of chemical and physical properties were performed with GLM procedure of SAS (SAS, 2001) and Duncan Multiple Range Test were used to test differences between treatment.

## Results and Discussion

Values of qualitative parameters of meat in longissimus dorsi et lumbarum muscle of Rex and Satin Rabbits in the content of moisture ( $72.91 \pm 2.28\%$  vs.  $71.88 \pm 0.65\%$ ), crude protein ( $21.32 \pm 2.06\%$  vs.  $21.89 \pm 0.64\%$ ), crude fat ( $2.09 \pm 0.71\%$  vs.  $2.00 \pm 0.58\%$ ), energy ( $4769.00 \pm 45.14$  kkal/kg vs.  $4815.67 \pm 133.81$  kkal/kg). Result showed that there were no differences between breed of rabbits on chemical properties. Moisture of Rex and Satin rabbits were  $72.91 \pm 2.28\%$  and  $71.88 \pm 0.65\%$ , respectively. The similar result of moisture of rabbit meat were reported 74.9% on lower growth rabbit, 75.1% on control and 75.3% on fastgrowing rabbit (Gondret *et al.* 2005). Mertin *et al.* (2012) reported moisture of hare and rabbit were 72.48% and 74.25%. Crude protein content of Rex and Satin rabbit meat were similar with other study, 22.20% (Mertin *et al.*, 2012), 20.8% (Lebas *et al.* 1986), lower on local rabbit was  $18.36 \pm 0.58\%$  (Brahmantiyo *et al.* 2014). Crude fat content were similar with Mertin *et al.* (2012) that reported on rabbit and hare were 2.55% and 1.23%, respectively, lowergrowth, control and highergrowth rabbits were 2.1%, 2.3 and 2.5%, respectively (Gondret *et al.* 2005) and lowest on local rabbit was  $1.34 \pm 0.64\%$  (Brahmantiyo *et al.* 2014). Energy content in Rex and Satin rabbits were higher than local rabbit with  $1860.67 \pm 1310.00$  kkal/kg (Brahmantiyo *et al.* 2014) and domestic rabbit with 468.01 kJ/100 g (Mertin *et al.* 2012).

Table 1. Chemical properties of rabbit meat

Traits	Breed	
	Rex	Satin
Moisture (%)	$72.91 \pm 2.28$	$71.88 \pm 0.65$
Crude protein (%)	$21.32 \pm 2.06$	$21.89 \pm 0.64$
Crude fat (%)	$2.09 \pm 0.71$	$2.00 \pm 0.58$
Energy (KKal/kg)	$4769.00 \pm 45.14$	$4815.67 \pm 133.81$

Chemical properties of Rex and Satin were not affected by breed, Judge *et al.* (1989) explained that breed, age, species, location of muscle and nutrient content of feed is very influential on the chemical properties of the meat. Rex and Satin rabbits were same type of medium rabbit, which produced fur and meat, and in this study they were kept in the same condition (feed and environmental).

At industrial level, ultimate pH is the main parameter used to measure meat quality (Maria *et al.* 2006). Lawrie (2003) stated that decreasing of meat pH caused accumulation of lactic acid after slaughter. No differences in meat pH were detected between breeds and location of muscles. Meat pH were ranged between  $5.58 \pm 0.48$  to  $5.82 \pm 0.16$  and meat in this conditions were generally similar with Ultimate pH of animal (Soeparno, 1992; Blasco *et al.* 1992). Gondret *et al.* (2005) obtained a value of ultimate pH in rabbits with a slow growth rate, control and fastgrowth at 5.74 and Zeferino *et al.* (2012) obtained a pH value of 5.92 and 5.90 in the 24-hour measurement after slaughter, and amounted to 5.93 and 5.95 in the 48-hour measurement after slaughter. WHC of longissimus dorsi et lumbarum muscle of Rex rabbit was 65.52

$\pm 14.73$  mg H<sub>2</sub>O, lowest than biceps femoris muscle of Rex was  $85.21 \pm 2.75$  mg H<sub>2</sub>O, Biceps femoris muscle of Satin was  $84.76 + 14.27$  mg H<sub>2</sub>O and Longissimus dorsi et lumbarum muscle of Satin was  $82.72 + 3.98$  mg H<sub>2</sub>O). Ultimate pH affect WHC indirectly (Maria et al. 2006).

Table 2. Physical properties of rabbit meat

Traits	Rex		Satin	
	<i>Biceps femoris</i>	<i>Longissimus dorsi et lumbarum</i>	<i>Biceps femoris</i>	<i>Longissimus dorsi et lumbarum</i>
pH	$5.58 \pm 0.48$	$5.72 \pm 0.17$	$5.82 \pm 0.16$	$5.73 \pm 0.13$
Water holding capacity (mg H <sub>2</sub> O)	$85.21 + 2.75^a$	$67.52 + 14.73^b$	$84.76 + 14.27^a$	$82.72 + 3.98^a$
Tenderness (WBshear)	$1.41 + 0.16^a$	$1.41 + 0.31^a$	$1.65 + 0.38^a$	$1.11 + 0.28^b$
Cooking loss (%)	$32.60 + 10.05$	$32.64 + 10.79$	$37.12 + 6.53$	$32.16 + 9.92$

Tenderness of meat of longissimus dorsi et lumbarum muscle of Satin rabbit were very tender than others. Expressing of satisfaction of eating meat result from the interaction of this quality characteristic with other factors like juiciness and flavor (Koomarie, 1996). Maria *et al.* (2006) reported tenderness of rabbit meat that were slaughtered in summer and winter had shear force value as  $0.61 \pm 0.04$  and  $1.04 \pm 0.03$ . Brahmantiyo *et al.* (2014) reported higher tenderness of Rex with shear force value were  $4.54 \pm 0.13$  on male and  $4.44 \pm 0.30$  on female and Zeferino *et al.* (2012) obtained tenderness of purebred and crossbred rabbits were 2.64 and 2.70, respectively.

Cooking loss of Rex, Satin with different muscle location were no difference. Yalcin *et al.* (2006) obtained cooking loss of male and female rabbit were 39.31 (0.45) and 38.14 (0.68), Zeferino *et al.* (2012) reported cooking loss of purebred and crossbred were no differences (35.59 vs. 35.52). Cooking loss of the meat were similar to those obtained by Hernandez *et al.* (1998) and Pla *et al.* (1998). These differences might be due to age, slaughter age and slaughter weight of rabbits.

## Conclusion

Rex and Satin rabbits were known as pet animal that had ability to produce fur and meat. Rabbit meats from Rex and Satin were had good enough in quality such as chemical and physical traits and similar with broiler rabbits.

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# Propolis, Pollen, and Honey Production on Two Different Agroecosystem

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## Abstract

*Trigona spp.* is a stingless bee which is not cultivated yet, because it produced less honey than other genus but *Trigona* is a good propolis producer. The aim of this study was to study propolis, pollen, and honey production in two different Agroecosystem. Six colony were cultivated in two Experimental Plantation (Cicurug Monoculture Agroecosystem) and Community Plantation (Cijeruk Polyculture Agroecosystem), and directly observed for propolis, pollen and honey production. The results showed that propolis produced at monoculture agroecosystem is 27,79 g, and 48,80 g at polyculture agroecosystem. Pollen that produced at monoculture agroecosystem is 30,20 g and at polyculture agroecosystem is 43,25 g. *Trigona* at monoculture agroecosystem produced honey 7,58 g, but at polyculture agroecosystem only 0,70 g. The products that produced by *Trigona* at two different agroecosystem, influence by flowering season, distance between plant and the colony, and environment.

**Keywords:** agroecosystem, honey, pollen, propolis, trigona

## Introduction

*Trigona spp.* is a stingless bee with a body size between 3 – 8 mm and very dynamic. *Trigona* can be detected by their way of life, outside and inside the nest. *Trigona* is less cultivated than other genus.. Stingless bee contributes to preservation of biodiversity by conserving populations of rare plants species. The colonies rarely abscond and resistant to the diseases and parasites of honey bees (Heard, 1999). Honey bee can be breed productively throughout the year in tropic area because plants as food sources are available continously. Beekeeping should be close to the flowering plants such as plantations or forests to gain higher productivity. *Trigona* and *Apis* are opportunist organisms, so all kinds of plants can be used as their food sources. Pollen is rich in proteins, vitamins and minerals and provides these nutrients to the bees. Pollen may be packaged and used as food supplements and also added to infant food, also used in many cosmetics preparations. *Trigona* honey difficult to extract, but it produces more propolis (Singh, 1962; Kwapong *et al.*, 2010). *Trigona* propolis frequently used as natural medicine for healthcare and body resistance. Propolis is the resin collected by honey bee from plants bud and broken branch, mixed with enzyme from bee saliva and used to protect their nest from contamination of bacteria, virus and fungi (Ghisalberti 1979; Gojmerac 1983; Marcucci 1995; Popova *et al.* 2005; Chen *et al.* 2008). The aims of this study was to analyze the production of propolis, pollen and honey which were cultivated in two different Agroecosystem.

## Materials and Methods

Three colonies of *Trigona spp.* cultivated in monoculture agroecosystem and three colonies in policulture agroecosystem. Each colony consisted of  $\pm$  300 bees, including one queen, few drones, and hundreds of workers cultivated in stup made of wood. There were 437 nutmeg trees (*Myristica fragrans* Houtt) in monoculture agroecosystem. 10 nutmeg trees, 1 white leadtree, 1 dogfruit tree, 1 lengkeng tree, 2 mango trees, 4 the rambutan trees, 2 rose apple trees, 3 pineapple trees, 1 passion fruit tree, 2 guava trees, 1 am-barella tree, 3 durian trees, 5 papaya trees, 41 banana trees, 1 mangosteen tree, and 3 jackfruit trees in polyculture farm.

Stups made of jengjeng woods with of 25x15x15 cm<sup>3</sup> and were numbered at the front side, then weighed for the empty weight. All the brood, food, and bee colony were moved from the bamboo into the stup and also propolis from the old entrance onto the new entrance. The stup filled with the colony were weighed, then placed at the old site for about 3 days for adaptation and cultivated for 3 months. After that, the colony were moved to the research location.

The method used in this study was direct observation on production of *Trigona* cultivated at two different agroecosystem. The purposive sampling were used to determine the location based on the potential of *Trigona* plant, i.e. Experimental Plantation (Cicurug Monoculture Farm) built by BALITRO which is a nutmeg unit production at 550 m above sea level on A climate zone. Community Plantation (Cijeruk Polyculture Farm) with various food potential at 400 m above sea level and A climate zone.

The observed variables were the propolis, pollen and honey production. After cultivated, the whole harvested products were weighed, then we separated honey, pollen and propolis. Each type of product (Propolis, Pollen and Honey) were weighed separately. The resulting datas of the research would be descriptively explained using tables and figures.

## Results and Discussion

### Productivity of Colony

The colonies initial weight in polyculture was havier than monoculture, but after one week, the colonies in monoculture were always havier than in polyculture (Figure 1). The t-test showed that average colony weight gain in two agroecosystem were significantly different ( $P < 0,05$ ) with score  $95,5 \pm 10,8$  in monoculture and  $61,3 \pm 11,7$  in polyculture. The variety of colony weight was infected by flowering season and environment.

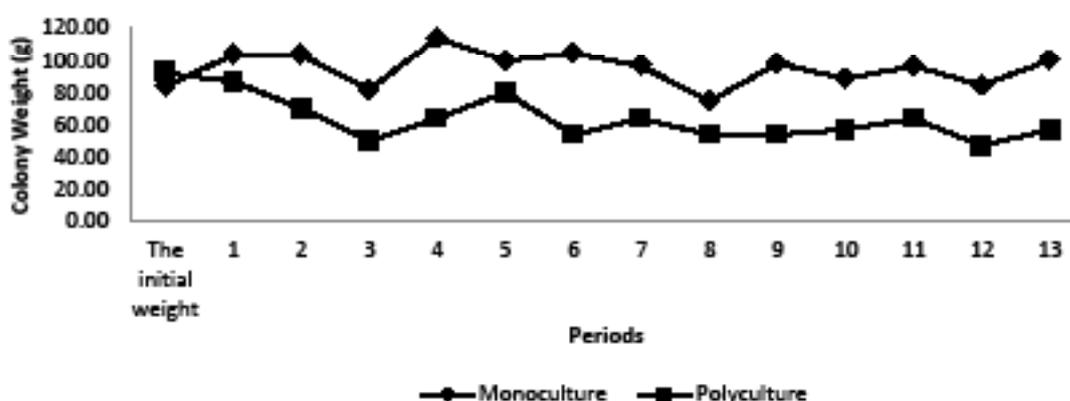


Figure 1. *Trigona* spp. Colonies Weight in Two Agroecosystem Farm

The flowering period coincides with the research period, thus results in colony growth. Honey bee needs food to fulfill their needs and colony growth. At the end of research period, the colony weight in monoculture was 299 g and polyculture 170 g. However, after harvesting, it turned out that *Trigona* in monoculture only produced 21, 93% from their colony weight, and 54, 56% on polyculture. The highest product of *Trigona* at monoculture was pollen, and propolis at policulture (Table 1). Perum Perhutani (1986) said, while building a nest, the bees will be very active to manufacture the cells as long as the forage and environments are in good condition, especially on flowering season. The weight gain in Monoculture was 80 % for brood cell and the product only 20%. Honey, Pollen and Propolis lower because they used to build the brood cell. The weight gain in Polyculture was lower but pollen, honey and propolis is 50% of the weight gain. Sihombing (1995) said, the presence of the queen affects pollen collecting through the egg-laying activity that will hatch. The smell of hatch will stimulate pollen collection. It will last simply until the maximum need of the colony fulfilled, so the storing of pollen in the nest will not exceed the colony needs for a certain period. Honey bee eats pollen as protein and fat source, also to nurse the brood hence the lower pollen production in monoculture. Kwapong *et al.* (2010) said, that stingless bees use cerumen (a mixture of wax and plant resin) in the construction of the brood cells and storage pots.

Nutmeg (*Myristica Fragrans* Houtt) is the plant that flowering throughout the year in Monoculture, but the high flowering period is on April and May. Colony growth at polyculture agroecosystem was less than at monoculture, but they made higher amount of product. It happened because during the research, flowering seasons only occurred on few plants, i.e. 10 nutmeg trees, 1 lamtoro tree, 3 jackfruit trees, 3 pineapple trees, 5 papaya trees, and 2 guava trees. Based on Siregar *et al.* (2011), *Trigona* needs the environment with vegetation that provide natural pollen and natural nectar in order to breed and produce a variety of bee products, such as honey, pollen and propolis. Allegedly, the amount of food collected in

two agroecosystem were used for colony growth, but the colony was not grow rapidly during the research, affected by unsupported environmental condition due to weather changes, that the rain is more often than the heat. The rainfall was high in March and decreased on April. On May, the rainfall raised up which greatly affected the colony growth. Sommeijer *et al.* (1983) said, foraging dan food resources was kept by the rain, because pollen resources was postponed by the rain in the morning and the continous rainfall will decreased foraging activitiessr because light intensity also decreased.

Table 1. Harvesting of 6 colony *Trigona* spp. on Monoculture and Polyculture Nutmeg Plantation

Agroecosytem	Final Colony Weight (g)	Harvest (g)/(%)	Propolis (g)/(%)	Pollen (g)/(%)	Honey (g)/(%)
Monoculture	299	65,57/(21,93)	27,79/(9,29)	30,20	7,58
Policulture	170	92,75/(54,56)	48,80/(28,71)	43,25	0,70

Source: Primary Data, 2012

Colony growth in monoculture were different from polyculture agroecosystem as shown in Figure 2. It showed that in the beginning of research, food supplies cells and brood cells in monoculture were nearly balanced, but after 3 months the colony were grown and the food supplies were low (Figure 2A - 2B). Otherwise, Figure 2C showed that food supplies and propolis were plenty than brood cells in polycultutr colonies at the beginnings, but after 3 months, the food supplies and brood cells were almost balance (Figure 2D).

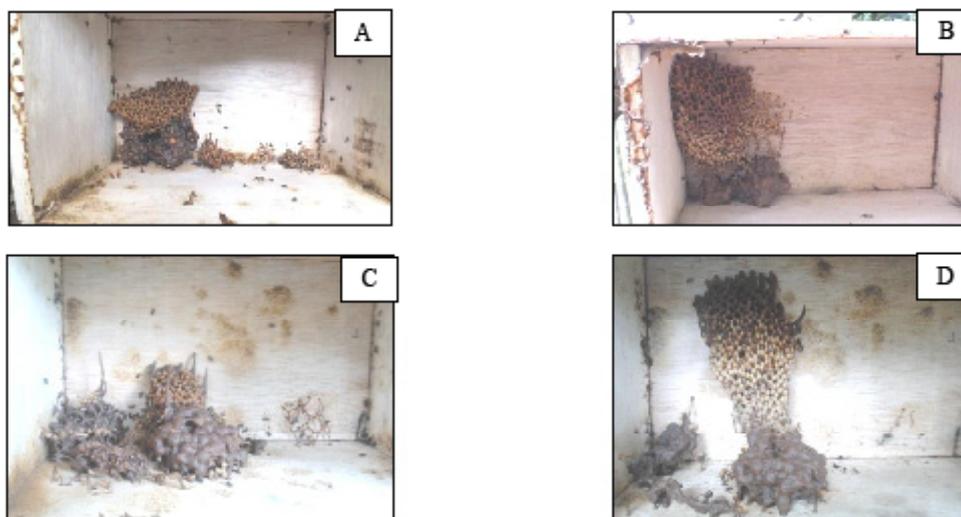


Figure 2. Colony Growth of *Trigona* spp. Two Agroecosystem. A). Colony of *Trigona* at Monoculture Agroecosystem on the beginning; B). Colony of *Trigona* at Monoculture Agroecosystem at the end of research; C). Colony of *Trigona* at Polyculture Agroecosystem on the beginning; D). Colony of *Trigona* at Polyculture Agroecosystem at the end of research

## Conclusion

Colony cultivating in two agroecosystem produced different amount of product. Colony in Monoculture produced 27,9 g Propolis and 48, 80g at Polyculture. Pollen produced in Monoculture is 30, 20gr and 43, 25 gr at policulture. Honey in Monoculture is 7,58 g, while Policulture is 0,70g

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# Distribution of Thermal Body Surface Ettawah Grade in Different Tropic Microclimates

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## Abstract

The purposes of this research were to inventory and compare means thermal body surface distribution of Ettawah Grade in tropic area with different microclimates. The research was conducted in Bogor district at Desember 2014. Purposive sampling method was used in this research. Eighth lactation Ettawah Grade was used. Fourth lactation Ettawah Grade were measured in elevation 210 MSL with average temperature (T)  $29.05 \pm 1.98^{\circ}\text{C}$ , relative humidity (RH)  $80.07 \pm 7.60\%$  and thermal humidity index (THI)  $81.50 \pm 2.41$ . Fourth Ettawah Grade were measured in elevation 675 MSL with average temperature (T)  $27.95 \pm 1.92^{\circ}\text{C}$ , relative humidity (RH)  $76.49 \pm 9.07\%$  and thermal humidity index (THI)  $79.14 \pm 2.10$ . Thermal body surface was measured using Flir Thermal Cam. Thermal body surface was classified into fifth major area's namely total body area, feet and legs area, chest and abdominal area, neck area and head area. Thermal body surfaces were measured in tenth hours start from 07.00 AM to 16.00 PM. The data's were analysed using descriptive statistic and compare means independent-sample T test. The results were indicated that average thermal of total body area is  $34.55 \pm 0.36^{\circ}\text{C}$  (210 MSL) and  $33.62 \pm 0.49^{\circ}\text{C}$  (675 MSL), feet and legs area is  $33.60 \pm 0.27$  (210 MSL) and  $32.16 \pm 0.44^{\circ}\text{C}$  (675 MSL), chest and abdominal area is  $34.78 \pm 0.43^{\circ}\text{C}$  (210 MSL) and  $34.09 \pm 0.50$  (675 MSL), Neck area is  $35.30 \pm 0.76^{\circ}\text{C}$  (210 MSL) and  $34.45 \pm 0.76$  (675 MSL), head area is  $35.66 \pm 0.70$  (210 MSL) and  $34.62 \pm 0.45$  (675 MSL). Compare means were showed that average thermal of whole body area, feet and legs area, and head area were significantly difference ( $P < 0.05$ ). The conclusion was obtained that warmest thermal body surface distribution is head area.

Keywords: ettawah grade, microclimates, thermal distribution

## Introduction

Microclimates are being strong related to the thermal body of ettawah grade. Thermal body of Ettawah Grade can be shown many things. System organ body always give a response for different condition of microclimates. In the condition microclimates are below to the comfort zone Ettawah Grade will get cold stress. In the night condition if temperature drops below  $21^{\circ}\text{C}$  for 3-6 h, the animal has sufficient opportunity to lose at night all the heat gained from the previous day (Igono, 1992; Silanikove, 2000). Otherwise in the condition microclimates are above to the comfort zone Ettawah Grade will got heat stress. Short-term preslaughter transport at high ambient temperatures may cause significant responses in goats (Kadim *et al.*, 2006). In mammals, the body temperature is maintained at a relatively constant level because of the balance that exists between heat production and heat loss (Silanikove, 2000). Thermal body of ettawah grade are produced from biologically process in the body to ensure vital organ work properly. Thermal body Ettawah Grade also can be used for biologically alarm in goat's soreness. Milk yield in dairy goats decreased as THI value increased, and for each 1 unit increasement of THI there is a decrease of 1% in milk yield (Salama *et al.*, 2014). In addition, uttermost scenarios of climatic change will negatively affect the dairy industry and that the importance of goats to the dairy industry will increase in proportion to the severances of changes in environmental temperature (Silanikove *et al.*, 2015).

In the recent years, the study about thermal body always measured in the rectal area. Rectal temperature is an indicator of thermal balance and may be used to assess the adversity of the thermal environment that can affect the growth, lactation, and reproduction of farm animals (Silanikove, 2000). Rectal temperature of the saanen goat was measured in turkey is  $39.0 \pm 0.03^{\circ}\text{C}$  (Keskin *et al.*, 2006). Futher more, rectal temperature of the crossbreed goat was measured in turkey is  $38.9 \pm 0.01^{\circ}\text{C}$  (Keskin *et al.*, 2006).

## Materials and Methods

### Material

Eighth lactation Ettawah Grade was used in these research. Fourth lactation Ettawah Grade were measured in middleland elevation (210 MSL) and Fourth lactation Ettwah Grade were measured in highland elevation (675 MSL). Research was conducted in Desember 2014 at Bogor district, West Java Provinces, Indonesia.

### Method

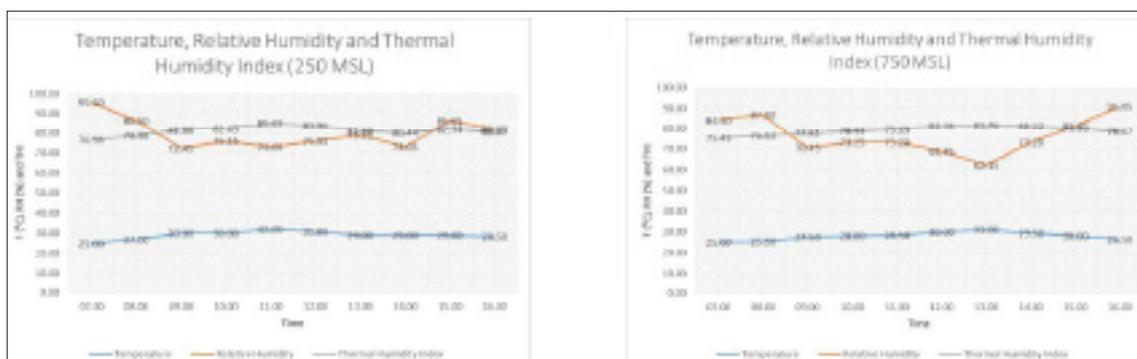
Purposive sampling method was used in this research. Microclimates data were collected in two areas. First area was collected in Dramaga regencies with elevation 210 MSL. Second area was collected in Taman Regencies with elevation 675 MSL. Microclimates data were collected are temperature and relative humidity. Temperature ( $^{\circ}\text{C}$ ) was measured using dry bulb thermometer. Relative humidity (%) were measured using digital hygrometer. In the current review the THI was calculated as follows:  $\text{THI} = (1.8 \times \text{Tdb} + 32) - [(0.55 - 0.0055 \times \text{RH}) \times (1.8 \times \text{Tdb} - 26.8)]$ , where Tdb is the dry bulb temperature ( $^{\circ}\text{C}$ ) and RH is the relative humidity (%) (Salama *et al.*, 2014).

Thermal body surface was measured using Flir Thermal Cam. Thermal body surface variables were measured classified into fifth areas namely whole body area, feet and legs area, chest and abdominal area, neck area and head area. Thermal body surface was measured from the side view. The data was tabulated based on fifth categories. The data were analysed using descriptive statistic and compare means independent-sample T test.

## Results and Discussions

### Temperature (T), Relative Humidity (RH) and Thermal Humidity Index (THI)

On the whole temperature in the two areas with elevation 210 MSL (Graph 1) and 675 MSL (Graph 2) slight similar. There was similar pattern in the beginning to the end. At 07.00 AM temperature in two areas are  $25^{\circ}\text{C}$ . Furthermore, temperature slight increase in the both of area. In the area with elevation 210 MSL had peak temperature at 11.00 AM, afterward slight decrease. Otherwise the area with elevation 675 MSL had peak temperature at 13.00 PM, afterward slight decrease. The temperature at 16.00 PM between 250 and 675 MSL was different. There are  $28.5^{\circ}\text{C}$  (210 MSL) and  $26.5^{\circ}\text{C}$  (675 MSL). The average temperature in tropic area is  $24\text{--}34^{\circ}\text{C}$  with relative humidity between 60-90% (Yani *et al.*, 2007).



Graph 1. Temperature, relative humidity and thermal humidity index (210 MSL)

Graph 2. Temperature, relative humidity and thermal humidity index (675 MSL)

The relative humidity in the area with elevation 210 MSL had been peak at 07.00 AM (95.60 %). Afterward slight decrease to 09.00 AM (72.45 %). That is the lowest level. Furthermore, relative constant to 14.00 PM. In the area with elevation 675 MSL had been peak relative humidity at 16.00 PM (91.05 %) and lowest level at 13.00 PM (62.15 %).

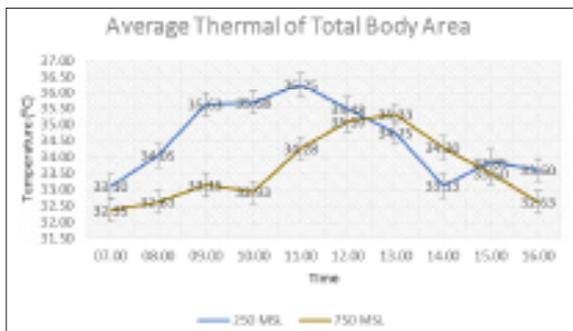
Table 1. average temperature, relative humidity and thermal humidity index (THI)

Elevation (MSL)	Temperature ( $^{\circ}\text{C}$ )	Relative Humidity (%)	Thermal Humidity Index
250	29.05 $\pm$ 1.98	80.07 $\pm$ 7.60	81.50 $\pm$ 2.41
675	27.95 $\pm$ 1.92	76.49 $\pm$ 9.07	79.14 $\pm$ 2.10

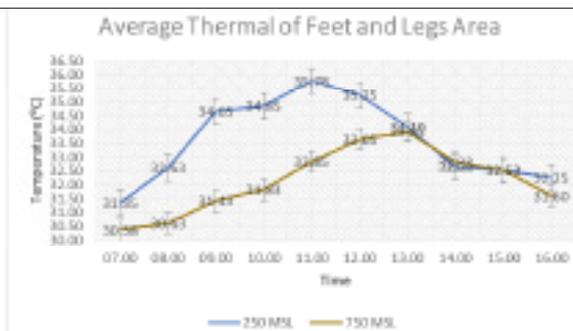
The average temperature between 07.00 AM to 16.00 PM in two was different 2.90°C. Furthermore, the average relative humidity was different 3.58 %. The thermal humidity index was different 2.36. On the whole area with elevation 675 MSL are lower than 210 MSL.

### Thermal Distribution of Total Body, Feet and Legs, Chest and Abdominal, Neck, Head

On the whole average thermal of total body area slight different between 210 MSL and 675 MSL. The area with elevation 675 MSL are lower. The average thermal of total body area is 34.55±0.36°C (210 MSL) and 33.62±0.49°C (675 MSL). The peak thermal is 36.25°C at 11.00 AM (210 MSL) and 35.33 at 13.00 PM (675 MSL). In the 12.00 PM temperature total body area in the 675 MSL is exceeded 210 MSL. After 13.00 PM sharp decrease.

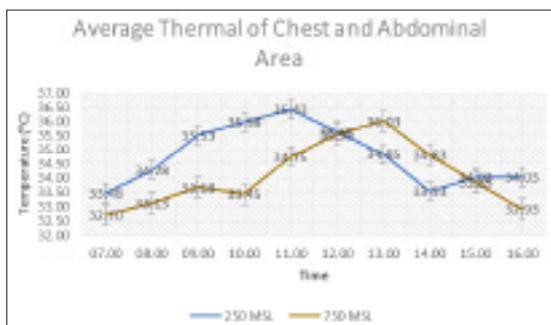


Graph 3. Average thermal of total body area

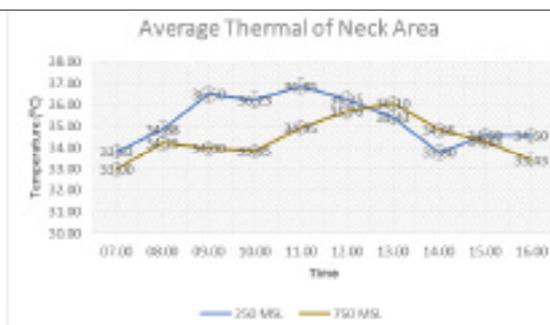


Graph 4. Average thermal of feet and legs area

The average thermal of feet and legs area pattern is slight similar with average thermal pattern of total body area (Graph 4). The average thermal of feet and legs area is 33.60±0.27 (210 MSL) and 32.16±0.44°C (675 MSL). The maximum temperature is 35.78°C at 11.00 AM (210 MSL) and 33.93°C at 13.00 PM (675 MSL). On the whole average thermal of feet and legs area is lower than average thermal of total body area 0.95±0.67°C (210 MSL) and 1.46±1.03°C (675 MSL). The average thermal of feet and legs area in the area with elevation 675 MSL is lower 1.44±1.04°C than area with elevation 210 MSL.



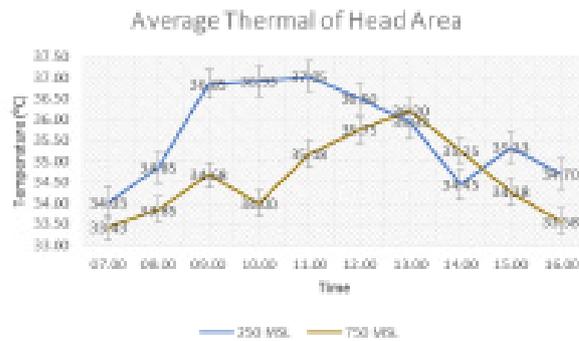
Graph 5. average thermal of chest and abdominal area



Graph 6 average thermal of neck area

The average thermal of chest and abdominal area trend is slight similar with average thermal of total body area trend (Graph 5). The average thermal of chest and abdominal area is 34.78±0.43°C (210 MSL) and 34.09±0.50 (675 MSL). The maximum temperature is 36.43°C at 11.00 AM (210 MSL) and 36.03°C at 13.00 PM (675 MSL). On the whole average thermal of chest and abdominal area is higher than average thermal of total body area 0.22±0.17°C (210 MSL) and 0.47±0.33°C (675 MSL). The average thermal of chest and abdominal area with elevation 675 MSL is lower 0.69±0.84°C than area with elevation 210 MSL.

The average thermal of neck area graph is sharp different with average thermal of total body area graph (Graph 6). The peak temperature is 36.85°C at 11.00 AM (210 MSL) and 36.10°C at 13.00 PM (675 MSL). On the whole average thermal of neck area is higher than average thermal of total body area 0.75±0.55°C (210 MSL) and 0.83±0.59°C (675 MSL). The average thermal of neck area with elevation 675 MSL is lower 0.85±0.84°C than area with elevation 210 MSL.



Graph 7 average thermal of head area

The average thermal of head area pattern is sharp different with average thermal of total body area pattern (Graph 7). The peak temperature is 37.05°C at 11.00 AM (210 MSL) and 36.20°C at 13.00 PM (675 MSL). On the whole average thermal of head area is higher than average thermal of total body area  $1.11 \pm 0.78^\circ\text{C}$  (210 MSL) and  $1.00 \pm 0.71^\circ\text{C}$  (675 MSL). The average thermal of head area with elevation 675 MSL is lower  $1.04 \pm 0.89^\circ\text{C}$  than area with elevation 210 MSL.

### Compare Means Independent-Sample T Test

Table 2 showed that compare means between area 210 MSL and 675 MSL had been result significant different in average thermal of total body, feet and legs, neck area and head area ( $P < 0.05$ ). The abdominal and chest area were showed insignificant different.

Table 2 Compare means average thermal in different elevation area

Elevation Area	Total Body Area	Feet and Legs Area	Abdominal and Chest Area	Neck Area	Head area
210 MSL	0.022	0.01	0.082	0.165	0.046
675 MSL	0.024	0.03	0.083	0.165	0.053

### Conclusion

The conclusion of distribution thermal body surface in different microclimates is the warmest area in head area.

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# Development Strategies of Community Dairy Farms in Karo Regency, North Sumatera

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## Abstract

*Dairy-cattle farmings with small scale system in Karo Regency-North Sumatera are commonly practiced by local farmers as one of the livestock activities which have significant contributions to farmers income. Some biological and social constraints were found to be limiting factors affecting the productivity and benefits to small farmers. This study was aimed at analysing the internal and external factors influencing animal production including productivity, the availability of forages for animal feeding, farmers and group capacity, technology and other related socio-economic aspects using SWOT analyses. Survey method involving farmer respondents was used and observation was made to the location of cattle farming to identify prospects and problems faced by farmers and provide some alternatives for solution. The results showed that based on the score and weight counted from the internal factors, of 2.669 and external factor of 2.525, indicating that the dairy production in Karo Regency, was categorized as “developing production status”. Taking into consideration the levels and the factors involved, and supporting aspects in all levels of production, an integrated approach would be one of the alternative to improve production through the optimisation use of local resources including human and natural resources.*

*Keywords: dairy cattle, farmer, Karo-Regency, North Sumatera*

## Introduction

Dairy farming industry plays a crucial role in the efforts to meet such human food needs as milk, butter, evaporated milk, ice cream, cheese, and dried milk products. Developing this industry means encouraging farmers to develop their dairy farms. In fact, there have been a great number of dairy farms in Indonesia. However, nationally the milk production is still far below the needs. According to Directorate General of Livestock (2010), 80% of the national demand for milk was still supplied by milk imports. Undoubtedly, this condition should provide an excellent opportunity for local farmers to increase the population and productivity of their dairy cows to reduce dependence on imports. Based on business scales, dairy farms can be classified into community dairy farms and dairy enterprises.

Most dairy farms in Indonesia are the family-size small farms, very often showing no sign of developing a sustainable system. Sarpintono (2013) said that even though the number of the dairy cows on lactation ranged from 3-4 cows per household farmer, the milk production has not been able to meet the national milk consumption and to increase dairy farming on a competitive basis. In order to succeed in dairy farming, a farmer should be able to combine the following elements: good management, good location for breeding, proper livestock composition, high producing cows, use of proper equipment, fertile land for forage crops, and good marketing (Sudono, 1999).

One of dairy farming areas in North Sumatra is Karo-Regency, which is located at an altitude of 280-1420 m above sea level. With temperatures ranging from 16.4°C-23.9°C, Karo-Regency is quite suitable for dairy farming. Based on the data of Central Bureau of Statistics in 2014, there was a decrease by 35.46% in dairy cow population in Karo-Regency in the period 2009-2011, but in the period 2012-2013, the dairy cow population increased by 29.7% compared to the population number in 2011. In general, however, for the last five years, the data showed a population decline of 16.70%. In the meantime, the low productivity of community dairy farms is influenced by internal as well as external environments: dairy cow population, productivity level, scale of livestock ownership, forage production for livestock feed, human resources, land resources, people's purchasing power, development of science and technology, infrastructure and regional autonomy, and local government policy. In this connection, this study was carried out to formulate

and establish appropriate alternative development strategies in improving the productivity of community dairy farming in Karo-Regency.

## Materials and Methods

The research used expert respondents who were determined by purposive sampling: 2 academicians, 1 private party, and 1 officer from the Regency Husbandry Agency. The strategy preparation was carried out by classifying two factors, namely internal and external environments. Determination of the internal and external factors was done in several stages. The stages included data collection (identification of internal and external factors, determination of weight and rating), and the internal-external matrix analysis. SWOT matrix was made to formulate development strategies for community dairy farms.

## Results and Discussion

### General State of The Region

Geographically, Karo-Regency is located between 02°50' to 03°19' North Latitude and 97°55' to 98°38' West Longitude. Karo-Regency is one of the regencies in the development region of dairy cows in North Sumatra. Climatic suitability is the main supporting factor for the community dairy farms in Karo-Regency. Karo-Regency is surrounded by many regencies: to the north Karo-Regency borders is Langkat-Regency and Deli Serdang-Regency, to the east is Deli Serdang-Regency and Simalungun-Regency, to the south is Dairi-Regency and Toba Samosir-Regency, and to the west is Aceh Tenggara-Regency

### Identification of Internal Environment

The result of the internal environment identification showed a total weight score of 2.625, which means that the community dairy farming was in a strategic position and had enough strength to overcome weaknesses of the internal environment. The environmental strength which had the most influence on dairy farming was the availability of land with a value of 0.124. The availability of land made it possible for the expansion of cow housing and cultivation of forage that enabled to increase the scale of livestock raising. The internal environment weakness that had the most influence on dairy farming was lack of experience and skills in dairy farming. The internal environment of the community dairy farming in Karo-Regency, based on rating weight and weight score, is presented in Table 1.

Table 1. Internal factors of the community dairy farming in Karo-Regency

Internal Environment	Weight	Rating	Weight Score
<b>Strength</b>			
1. Availability of land	0.124	3	0.372
2. Availability of forages	0.112	3	0.336
3. Availability of seeds	0.109	3	0.327
4. Availability of water	0.109	3	0.327
5. Availability of business credit	0.072	3	0.216
6. Availability of labor	0.058	3	0.174
7. Potential of agricultural waste as feed	0.041	3	0.123
<b>Weakness</b>			
1. Experience and skills of farming	0.068	2	0.136
2. Availability of stall equipment	0.057	2	0.114
3. Availability of concentrate	0.054	2	0.108
4. Availability of medicines	0.05	2	0.1
5. Availability of technical training and extension	0.044	2	0.088
6. Availability of cooperative	0.04	2	0.08
7. The level of production and quality of milk	0.036	2	0.072
8. The level of milk processing	0.026	2	0.052
Total	1		2.625

Source: Primary Data (2015)

## Identification of External Environment

The result of the external environment identification showed a total weight score of 2,525, which means that the community dairy farming was in a strategic position and had enough opportunity to cope with the threat of the internal environment. The opportunity of the external environment that had the most influence on dairy farming was the natural and climatic conditions for the development of the community dairy farming with a value of 0.142. The threat of the internal environment that had the most influence on dairy farming was the interest in dairy cattle raising with a value of 0.167. Raising dairy cows in Karo was the people's lowest interest compared to other farm businesses. The external environment of the community dairy farming in Karo Regency, based on rating weight and weight score, is presented in Table 2.

Table 2. External factors of the community dairy farming in Karo-Regency

External Environment	Weight	Rating	Weight Score
<b>Opportunity</b>			
1. Natural conditions and climate for development	0.142	3	0.426
2. Support of government policies and programs	0.108	3	0.324
3. Availability of supporting infrastructure	0.1	3	0.300
4. Conditions of politics, law and security	0.09	3	0.270
5. Business competition	0.085	3	0.255
<b>Threat</b>			
1. Interest in raising dairy cattle	0.167	2	0.334
2. Awareness of the nutritional value of milk	0.109	2	0.218
3. Risks of livestock health problems	0.076	2	0.152
4. Support of milk processing industry	0.072	2	0.144
5. Mastery of Science and Technology	0.034	2	0.068
6. The attractiveness of other sectors	0.017	2	0.034
<b>Total</b>	<b>1</b>		<b>2.525</b>

Source: Primary Data (2015)

The result of the total score weight of the internal environment was 2.625 and the external environment 2.525. The position of the community dairy farm was in cell V of the external-internal matrix. Rangkuti (1999) stated that when the position of IE matrix is in cell 5, the suitable development strategy is a growth strategy through horizontal integration. The strategies that can be applied are the enlargement of raising business scale, market penetration by expanding the milk marketing, and product development by diversifying processed products. The alternative strategies include the strategies of S-O (Strength-Opportunity), that is, using the strength to take advantage of the opportunity; S-T (Strength-Threat), that is, using the strength to overcome the threat; and W-O (Weakness-Opportunity), that is, minimizing the weakness to make use of the opportunity.

### Strategy of S-O (Strength-Opportunity)

*Increasing the community dairy cow population through economic scales (S1, S2, S3, S4, S5, S6, S7, S8, O1, O2, O3, O4, O5).*

The community dairy cow population in Karo-Regency until now is relatively small, only 70 heads, with an ownership scale of less than 4 heads per breeder. To increase the population of dairy cows in Karo-Regency can be done by increasing access to small loans. The addition of dairy cow population is supported by the availability of land for the cultivation of forage and the construction of a new dairy cattle housing. The development of the community dairy farming through the population increase is possible by utilizing agricultural waste for concentrate making for cattle feed.

### Strategy of W-O (Weakness-Opportunity)

*Improving farmer technical capabilities through intensive training and extension (S5, S6, O1, O2, and O5).*

The average technical ability of the dairy farmers in Karo-Regency based on the principles of Good Dairy Farming Practices has not met a category of good (2.60). Increasing farmer technical capabilities is imperative, especially in the aspect of animal health, since most farmers lack this technical ability which includes knowledge of disease and disease prevention, where they are in a category of very poor. Improving

the quality of farmer resources should be supported by the ability and capacity of extension officers. Quddus (2013) said that the development of the community dairy farming should be in line with the increase capacity extension. Extension needs to be strengthened by the transfer of technology for the improvement of raising management to enrich farmer knowledge.

### **Strategy of S-T (Strength-Threat)**

*Increasing the empowerment program of the competitive processing dairy products (S5, S7, T2, and T3).*

Many farmers had constraints in the marketing of fresh milk. The direct marketing of fresh milk was carried out by the dairy farmers in Karo-Regency because they did not have a cooperative as the collector of milk from the farmers. The direct marketing of fresh milk put farmers in a difficult situation due to consumers' low level of demand and consumption for fresh milk, while at the same time the ability of farmers in processing fresh milk into various forms of processed products was also very limited. Increasing the empowerment program in milk processing should be done to encourage farmers to carry out diversification of processed products.

### **Strategy of W-T (Weakness-Threat)**

*Establishing a cooperative of dairy farmers in order to support the sustainability of the community dairy farming (S2, S3, S4, S5, O1, O2, O3, and O4).*

The availability of a cooperative for the community dairy farming in Karo-Regency is badly needed to support the sustainability of the dairy farming. The existence of a cooperative is expected not only to bring benefits to the farmers but also to overcome most of the problems facing dairy farmers in Karo-Regency in terms of the ease of milk marketing, the availability of feed, technical equipment and veterinary services, provision and distribution of credit facilities as a source of business capital, guaranteeing such risks as cattle death and business failure, the availability of complementary breeder needs and the provision of training and counseling to members. Urassa and Rafael (2002) said that the community dairy farming should be supported by the availability of a cooperative that serves as a milk collection place, provider of concentrate feed, medicine and counseling at an affordable cost for members.

## **Conclusion**

The development strategies of the community dairy farming that can be applied in Karo-Regency are growth strategies through horizontal integration such as the enlargement of raising business scale, market penetration by expanding the milk marketing, and product development by diversifying processed products. Alternative strategies based on SWOT analysis are: 1) increasing the community dairy cow population through economic raising scale, 2) improving the technical capabilities of farmers through intensive training and counseling, 3) increasing the empowerment program of the competitive processing dairy products, and 4) establishing a cooperative of dairy farmers in order to support the sustainability of the community dairy farming.

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# The Effect of Cage Floor Types on Growth Performance and Behaviour of Local Rabbit

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## Abstract

*This study aimed to identify production performance daily ingestive behavior of male local rabbit affected by different type of cage floor. Fifteen heads of local rabbit with initial weight  $0.82 \pm 0.07$  kg were used and allocated individually into different cage, namely bamboo floor (B), husk mats floor (H) and wire floor (W). The data of body weight gain, feed and water intake, daily behavior including ingestive, locomotion, elimination, grooming, resting behaviors were collected during 60 days. The behavior activity was recorded by using a scan sampling for ten minutes. The data of growth performance were analyzed with analyses of variance while the data of behavior were analyzed with non parametric Kruskal-Wallis. The results showed that the effect of different cage floors was not significantly different on all parameters with average of body weight gain  $12,5 \pm 1.3$  g  $h^{-1} d^{-1}$ . The rabbits were dominated by locomotion behavior during observation. Furthermore, between the three cage types were similar the eating patterns behavior observed, smelling, biting, chewing and swallowing. It can be concluded that the use of bamboo cage floor type, husks and wire can be used for raising rabbits by considering the availability of materials.*

*Keywords: behaviour, cage floor, growth, rabbit*

## Introduction

Rabbit is one of the animal that was still slightly contribute as animal food proteins source to people in Indonesia. Although this animal can utilize forage efficiently and rabbit meat is well known lower in fat and cholesterol content compared to other livestock. In general, rabbit intensively reared in a cage, so that the convenience of the cage is needed that would affect their productivity. One of the factors that influence it is the cage floor design. Comfort condition inside the cage reflects the welfare of livestock that should be considered by farmers in order to get maximum rabbit production level.

Some researches on the effect of type of cage floor on the productivity of rabbit were conducted by researchers. Siloto (2008) reported that rabbits were placed in a cage floored husk showed a positive impact on the welfare of rabbits compared with wire floors. Others research also conducted by Rashed and El-Edel (2015), Miko *et al.* (2012). Some researchers concluded that wire floor is unfavorable for rabbits, means that less welfare for the animals because the animals spent lower time for resting than others type of floor (Drescher 1992). Yet research on Indonesia local rabbit affected by type of floor cage is rarely done. Herman (2000) mentioned that the Indonesia local rabbits are more tolerant to heat (high temperature) than import rabbit. On the other hand, the design of the cage floor types will also affect the cost production in rabbit farming. Information on productivity and behavior of local rabbits that are kept on different types of floor cage is needed to be conducted. This study aimed to identify production performance and daily and ingestive behavior of male local rabbit affected by different type of cage floor.

## Materials and Methods

### Animals and Diets

The study was conducted in Laboratory of small ruminant production, Faculty of Animal Science, Bogor Agricultural University (IPB) Indonesia. Fifteen heads of local rabbit with initial weight  $0.82 \pm 0.07$  kg were used and allocated individually into different cage, namely bamboo floor (B), husk mats floor (H) and wire floor (W). Each treatment had three replication. The animals were fed with diet that was formulated with 19% DM of crude protein and formed as pellets (Table 1). *Ad libitum* watering was allowed to all animals during treatments. Individual feed intake were recorded daily, meanwhile behavior parameters were recorder weekly by scan sampling method for ten minutes. The data of body weight gain, feed and

water intake, daily behavior including ingestive, locomotion, elimination, grooming, resting behaviors were collected during 60 days.

Table 1. Chemical composition of diet (%)

	Nutrient					
	DM	CProtein	CFiber	CFat	Ash	NFE
Feed	88.12	19.13	20.09	3.37	9.66	47.75

### Statistical Analyses

The data of growth performance were analyzed with analyses of variance while the data of behavior were analyzed with non parametric Kruskal-Wallis (Steel & Torrie, 1980).

## Results and Discussion

### Feed Consumption

The dry matter intake of all animals in this trial were no significantly affected by type of floor cages ( $P>0.05$ ) with average daily intake 59 g. This means that different types of floor cage were not disturbing the consumption of rabbits, so that using bamboo that usually easy to handle by farmers did not negatively effect on feed consumption.

Table 2. Daily dry matter and nutrient intake of rabbit in different types of cage floor (g/h)

Parameter	Bamboo Floor	Husk Floor	Wire Floor	Average
Dry Matter	58.04±5.22	59.50 ±5.39	60.58±4.92	59.37±4.92
Crude Protein	11.08±0.97	11.31±1.04	11.53±0.94	11.31±0.22
Crude Fiber	17.52±1.56	17.97±1.64	18.30±1.49	17.93±0.39

### Growth Performance

Data of growth performance are presented in Table 3. It could be observed that the average daily body weight gain of all animals in this trial were no significantly affected by different of cage floor regime ( $P>0.05$ ).

Table 3. Production performance of rabbit in different type of cage

Parameter	Bamboo Floor	Husk Floor	Wire Floor	Average
Initial BW (g/h)	856±103	818±61	898±53	824±29
Slaughter weight	1502±117	1434±215	1625±93	1520±96
BWG (g/d/h)	10.54±2.13	11±3.03	13.04±3.81	11.53±1.33
FCR	5.64±0.9	5.69±1.3	5.04±1.7	5.46±0.4

Note: BWG (body weight gain), FCR (feed conversion ratio)

Table 2 shows that the average daily body weight gain of local rabbits in this study was relatively similar than the study conducted by Rashid (2009) and Noble (2010) that resulted  $13.88 \pm 1.60$  and  $12.63 \pm 0.63$  respectively. It means that in this study did not cause the negative effect for the animals. Indonesia local rabbit only can perform daily body weight gain at 10-20 g/h/d (Herman 2000).

### Behavior of Rabbit

Data of behavioral performance parameters are presented in Table 4. Despite, among treatments did not affected on behavior pattern of rabbit ( $P>0.05$ ), husk floor indicated rabbit had lower time for resting than other type of floor. This condition was relatively similar to that study conducted by Siloto (2008). This condition influence the grooming behavior that relatively indicated higher in husk floor, because rabbit need to groom more intense that caused by husk condition. Siloto (2008) also concluded that rabbit in a cage that was given husks do grooming more frequently than in a cage without husk.

Table 4. Behaviours frequency of local rabbit on different of cage floors (times/10 minutes)

Behaviour	Bamboo Floor	Husk Floor	Wire Floor
Feeding	2.00±0.41	2.18±0.65	1.95±0.59
Drinking	1.43±0.60	1.28±0.81	1.16±0.52
Eliminating	0.75±0.53	0.33±0.62	0.22±0.46
Grooming	1.91±0.95	2.11±0.49	2.07±0.44
Stereotyping	0.22±0.54	0.00±0.00	0.13±0.52
Resting	1.15±0.26	1.04±0.10	1.16±0.24

The others behavior also was not significant among treatment ( $P>0.05$ ). Behavior of urination in male rabbit is one of aggressive behavior. Male rabbits usually urinate to identify their domination to his rival (Cheeke *et al.* 2000). Fraser & Broom (2005) stated stereotype behavior is behavior that is done without a clear purpose and usually occurs in animals in the cage.

## Conclusion

The different types of cage floor was not influence on production performance and behavioral pattern of Indonesia local rabbit, even though husk floor indicated less favorable for rabbit in general. So that, it can be concluded that the use of bamboo cage floor type, husks and wire can be used for raising rabbits by considering the availability of materials.

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# Hycole and Hyla Rabbits Performance were Raised in Indonesia

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## Abstract

*Hycole was imported rabbit from France and Hyla was imported rabbit from China were raised at Indonesian Research Institute for Animal Production to evaluate their adaptability on tropical climate. Parent stock of Hycole (40 heads female line and 17 heads male line) and Hyla (40 heads female line and 20 heads male line) were used in these research. Data were collected of reproductive of does such as litter size at birth, litter size at weaned and mortality birth-wean and growth of rabbit such as weekly body weight of male and female from birth to 20 weeks of age. Gompertz non linear curve were used to evaluate body weight at mature and maturity ages of Hycole and Hyla. No differences between breed on reproductive traits of Hycole and Hyla, there were litter size at birth (8.1 + 2.4 kits vs. 8.1 + 2.0 kits), litter size at weaned (5.8 + 1.7 kits vs. 5.8 + 1.6 kits) and mortality (31.2 + 22.1 % vs. 30.4 + 21.1 %). Kits growth of Hycole since birth to five weeks of age were higher than Hyla, but no differences since six ages to 20 weeks of age. Estimated maturity of rabbit using Gompertz non linear curve were obtained Hycole rabbit was had age maturity lately than Hycole, on male (12.0 weeks vs. 10.7 weeks) and female (12.3 weeks vs. 10.3 weeks). HyCole and HyLa rabbits were had ability to develop as broiler rabbits in Indonesia.*

**Keywords:** Hycole, Hyla, litter size, mature age, rabbit

## Introduction

Rabbits development in Indonesia are increasing rapidly, especially in rabbit production centers that were stimulated growth of demand for rabbits. So far, farmers have not been able to breeding their rabbits so that there is a gap between demand and availability of rabbits. Rabbit market is very open with a profitable business, so availability is always lower than demand and this is the case in almost all developing area of rabbit.

Indonesian government were promoted rabbit meat as healthy food, because its had low cholesterol, low fat and high protein content. This promotion were developed market for meat from rabbit. Rabbit meat were produced from any kind of rabbit breed not especially from broiler rabbits because the farmer had no capability to produced broiler rabbits. Indonesian Research Institute for Animal Production (IRIAP) on 2012 and 2013, were imported broiler rabbits from France namely HyCole and from China namely HyLa, HyCole and HyLa were parent stock from male line and female line.

The broiler rabbits are mostly two- and four way crossbreds. The knowledges of the growth and development patterns of rabbits defined of genotype is the presumption of successful profitable production of meat animals (Vostry *et al.* 2008). In Indonesia, final body weight of rabbits for slaughter were above 2.00 kg and reached more than 6 months of age.

The research was undertaken to assess the litter and growth performance of an imported rabbits (HyCole and HyLa) under tropical condition of Indonesia.

## Materials and Methods

The research was conducted at Rabbit Laboratory, Indonesian Research Institute for Animal Production (IRIAP), Bogor, West Java. IRIAP is located at an altitude of 500 m above sea level with average ambient temperature between 20-30°C. The data generated for this study included 40 heads male HyCole Parent Stock ( $P_A$ ), 17 heads female HyCole PS ( $P_B$ ), 40 heads male HyLa PS ( $C_{AB}$ ) and 20 heads female HyLa PS ( $C_{CB}$ ). A total of 389 offspring of HyCole and 378 offspring of HyLa were evaluated from 48 mated HyCole and 49 mated HyLa.

Individual kit weight was taken at birth, 1 week to 20 weeks of age for both breeds. The weaning age was set at 35 days. Housing and feeding were similar in both breeds. Does and bucks were fed about 200-250 g/head on IRIAP formulated pelletized diet and watering ad libitum.

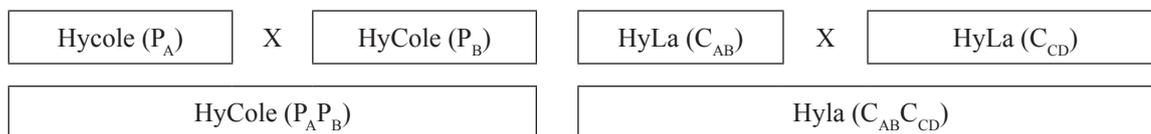


Figure 1. Mating diagram of HyCole and HyLa Rabbits.

Litter size at birth (LSB), litter size at weaning (LSW) and mortality during nursing period were evaluated as does performance. Body weight offspring at birth and weekly were weight until reached 20 weeks of age. Data were analyzed using procedure of general linear model (SAS, 2001). The Gompertz growth curve was fitted to the weight-age data of each rabbit by the nonlinear least squares procedure (SAS 2001). The equation for the Gompertz function has the following form:

$$y(t) = ae^{-be^{-ct}}$$

where  $y(t)$  is weight (in g) at age  $t$  (in days),  $A$ ,  $B$ ,  $K$  are fitted parameters, and  $e$  is the base of the natural logarithm ( $e=2.71828\dots$ ). Parameter  $A$  is interpreted as the asymptotic limit of the weight or the average mature weight of the rabbit,  $K$  is the rate of maturing (the larger the value of  $K$ , the earlier the animal matures), and  $B$  is a scaling parameter (constant of integration, related to the initial weight).

## Results and Discussion

Does performance of HyCole and HyLa were featured in Table 1. There no differences between HyCole and HyLa rabbits on LSB ( $8.1 \pm 2.0$  vs  $8.1 \pm 2.4$ ), LSW ( $5.8 \pm 1.6$  vs  $5.8 \pm 1.7$ ) and mortality during nursing ( $30.4 \pm 21.1$  vs  $31.2 \pm 22.1$ ). LSB and LSW of HyCole and Hyla rabbits were better than Rex (6.1 kits) and Satin (5.8 kits) rabbits that already raised in Indonesia since 1988 (Brahmantiyo *et al.* 2010). Akinsola *et al.* (2014) that raised Hyla in tropical condition in Nigeria showed that Hyla purebred were had litter size range between  $6.51 \pm 1.47$  to  $3.46 \pm 0.2$  kits and Hyla crossbred were had litter size range between  $6.24 \pm 0.63$  to  $2.79 \pm 0.38$  kits. Vaclavovsky *et al* (2000) reported litter size of 9.49 in HyLa purebred rabbits reared in Slovenia.

Table 1. Doe's performance

Parameters	HyCole (n = 48)	HyLa (n = 49)
LSB (heads)	$8.1 \pm 2.0$	$8.1 \pm 2.4$
LSW (heads)	$5.8 \pm 1.6$	$5.8 \pm 1.7$
Mortality (%)	$30.4 \pm 21.1$	$31.2 \pm 22.1$

Kits mortality during nursing period were still high, there were  $30.4 + 21.1\%$  for HyCole and  $31.2 + 22.1\%$  for HyLa. Olowofeso *et al.* (2012) reported mortality of crossbreeding between Rex (RX), Flemish Giant (GF) and Chinchila (CH) were reached 18.26 % on CHxGF to 30.03% on GFxRX that were raised in South West Nigeria.

Growth since birth to 20 weeks of age of HyCole and Hyla rabbits were featured in Table 2. HyLa rabbits were growth higher than HyCole on birth until 4 weeks of age and no differences between breed for the rest of growth. HyLa rabbits expressing mothering ability better than Hycole because on that stage of nursing period. HyCole and Hyla rabbits were reached slaughter weight on Indonesian standard on 14 weeks of age with body weight of  $2110.7 \pm 401.7$  g on HyLa and  $2111.9 \pm 462.3$  g on HyCole. These imported breeds were faster on growth than Rex and Satin rabbits which were had body weight at 14 weeks of age were  $1617.53 \pm 357.38$  g (female Rex),  $1581.59 \pm 326.60$  g (male Rex),  $1314.67 \pm 270.57$  g (female Satin) and  $1907.00 \pm 297.19$  g (male Satin) (Brahmantiyo *et al.* 2010). These Imported rabbit breeds were lower than Hyplus on 14 weeks of age which had  $2822.48 \pm 36.43$  g (Vostry *et al.* 2008).

Table 2. Rabbit growth

Body weight	HyLa (n = 378)	HyCole (n = 389)
Birth	57.8 ± 11.4 <sup>a</sup>	54.3 ± 11.4 <sup>b</sup>
1 week of age	145.3 ± 30.1 <sup>a</sup>	127.9 ± 33.1 <sup>b</sup>
2 weeks of age	223.9 ± 68.5 <sup>a</sup>	203.6 ± 56.4 <sup>b</sup>
3 weeks of age	336.3 ± 117.9 <sup>a</sup>	290.1 ± 109.7 <sup>b</sup>
4 weeks of age	469.5 ± 141.4 <sup>a</sup>	421.0 ± 130.9 <sup>b</sup>
5 weeks of age	663.5 ± 206.0	594.9 ± 167.8 <sup>b</sup>
6 weeks of age	731.1 ± 192.8	752.6 ± 161.4
7 weeks of age	940.1 ± 196.4	937.4 ± 204.6
8 weeks of age	1127.8 ± 288.7	1122.8 ± 249.4
9 weeks of age	1313.7 ± 357.4	1235.1 ± 286.0
10 weeks of age	1459.7 ± 373.1	1453.9 ± 324.9
11 weeks of age	1565.6 ± 388.4	1588.4 ± 371.5
12 weeks of age	1800.4 ± 384.4	1724.6 ± 399.2
13 weeks of age	1898.5 ± 371.9	1855.3 ± 402.7
14 weeks of age	2110.7 ± 401.7	2111.9 ± 462.3
15 weeks of age	2313.2 ± 387.0	2288.1 ± 495.1
16 weeks of age	2466.2 ± 425.8	2394.9 ± 539.2
17 weeks of age	2643.2 ± 399.1	2565.7 ± 502.9
18 weeks of age	2799.4 ± 445.1	2704.4 ± 470.9
19 weeks of age	2950.5 ± 452.7	2918.9 ± 441.8
20 weeks of age	3045.6 ± 451.7	3095.4 ± 434.0

Different superscript in the same line means significantly different (P<0.05).

**Growth curve**

Result of estimated equation of growth curve on Hyla (C<sub>AB</sub>C<sub>CD</sub>) and Hycole (P<sub>A</sub>P<sub>B</sub>) rabbits using Gompertz model were described below:

Y = 4 914.71 exp (- 3.73 exp -0.11t) for female HyLa (C<sub>AB</sub>C<sub>CD</sub>).....(1)

Y = 4 774.05 exp (-3.54 exp -0.10t) for male HyLa (C<sub>AB</sub>C<sub>CD</sub>) .....(2)

Y = 4 161.08 exp (-3.62 exp -0.12t) for female HyCole (P<sub>A</sub>P<sub>B</sub>) .....(3)

Y = 4 308.81 exp (-3.59 exp -0.12t) for male HyCole (P<sub>A</sub>P<sub>B</sub>) .....(4)

Graph of estimated growth curve using Gompertz model on HyLa and Hycole rabbit with different sex were figured below.

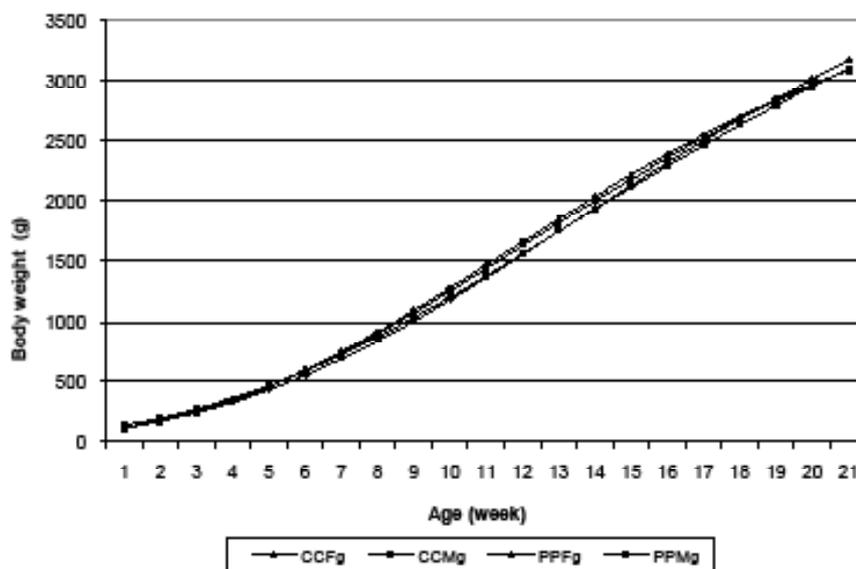


Figure 2. Estimated growth curve by Gompertz model, which CCFg for female HyLa, CCMg for male HyLa, PPFg for female HyCole and PPMg for male HyCole

On rabbits, Gompertz models can also be used to predict the inflection point of body weight and inflection point of age, as shown in Table 3.

Table 3. Prediction of inflection point of body weight and age of HyCole and Hyla with different sex.

Genotype	Breeds and sex	Inflection point of body weight (g)	Inflection point of age (weeks)
C <sub>AB</sub> C <sub>CD</sub>	Hyla female	2 256.2	12.3
C <sub>AB</sub> C <sub>CD</sub>	Hyla male	2 191.6	12.0
P <sub>A</sub> P <sub>B</sub>	Hycole female	1 910.2	10.3
P <sub>A</sub> P <sub>B</sub>	Hycole male	1 978.1	10.7

Inflection point is the point where the body weight of animal husbandry decreased growth rate in unit time inflection point of age or weight of animal reached puberty. Hyla male and female were reached maturity on 12.0 weeks and 12.3 weeks of age, respectively and much longer time than Hycole male and female who were reached maturity on 10.7 weeks and 10.3 weeks of age, respectively. Brahmaniyo *et al.* (2010) and Brahmaniyo and Raharjo. (2011) reported that Rex and Satin rabbits were reached maturity on 8.6 weeks and 10.3 weeks of age, respectively and selection changed maturity of Rex rabbits from 8.6 weeks became 10.1 weeks. On broiler rabbits, Dedkova *et al.* (1999) were reported that the crossbreds growth curve reaches the inflection point at the age from 7 to 10 weeks. Larzul dan de Rochambeau (2004) reported that selection on the growth rate will decrease the adult age when rabbit are slaughter at a predetermined weight (constant), so the slaughter weight can be achieved in a shorter time. He added that the selection process the growth rate effect on growth patterns of rabbits.

## Conclusion

HyCole and HyLa rabbits were well adapted in tropical condition in Indonesia and had LSB and LSW higher than Rex and Satin rabbits that were earlier imported to Indonesia. HyLa rabbits had higher potential on body weight than HyCole that were represented by inflection point on age. HyCole and HyLa rabbits were had ability to develop as broiler rabbits in Indonesia.

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# Constraints to, Challenges of, and Opportunities for Rearing Goats in Bali Province. A Case Study: Rearing Kids in Karangasem Regency

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## Abstract

*This study was undertaken to establish a database identifying constraints to, challenges of and opportunities for rearing goats in Bali Province. A survey was conducted on 63 goat farmers integrated with vegetable farms owning a total of 1,163 goats in Karangasem Regency. Data on growth performance and flock dynamics (birth, death, survival and sales) of 568 kids born in 2014 were collected between April and September 2014. 413 kidding occurrences delivered 568 kids; 65% were single born, 33% were twins and only 2% were triplets or quadruplets. The kids were a mixture of Etawah, Kacang, PE, Gembrong, Benggala and their crossbreds that had a relatively low mortality rate (8.6%), and litter size was on average 1.65 kids from does with 1<sup>st</sup> to 11<sup>th</sup> parity. Kids were housed in battery or colony systems and fed *Caliandra calothyrsus*, *Pennisetum purpureum* and 'dagdag' where cut and carry feeding systems were the only rearing system used. The average live weights of kids increased ( $P < 0.01$ ) from birth (3.1 kg) to a weaning weight of 18.2 kg at day 135. This growth was supported by the abundance of available feed for the does, and the housing used meant the incidence of parasites was low as indicated by FAMACHA<sup>®</sup> scores which were generally low ( $P < 0.01$ ). Of these goats 23 were sold for a total of IDR 35 M. High demand of 'Mecaru' kids for Hindu ceremonies, abundant water, free access to conservation forest for growing plants for roughage, assistance from Government Programs are all opportunities for increased goat production.*

**Keywords:** database, growth performance, integrated farming system, kids rearing, Mecaru

## Introduction

The current and potential role of the goat industry in Bali Province is not well understood and long-term research and development programs for goats reared by smallholder farmers in this province are poorly described. As a result, baseline quantitative information on goat farmers as well as the reproductive and productive parameters of their goats is also limited. On the other hand, goat production has the potential to alleviate and improve nutrition especially for women and children in rural areas (Devendra 2010) and to increase the income of smallholder goat farmers in Bali Province (Suciani *et al.* 2013).

Thirty-two million Indonesians out of a population of 234 million live below the national poverty line (BPS-Indonesia 2013) which is defined as living in extreme poverty below US\$ 1.25 a day in 2013 ([www.worldbank.org](http://www.worldbank.org)). In 2014 the goat density and the ratio of goats to humans was 11 goats/km<sup>2</sup> and 16 goats/1,000 people in Bali, respectively (BPS-Bali 2014). To help alleviate poverty as well as to improve nutrition of villagers, activities should be undertaken to improve the number of goats in Bali Province and more importantly to improve their productivity.

To improve the productivity of goats, attention should be given to important factors such as their genotype, the environment where goats are being raised, their live weight gain, and reproductive performance including rearing kids as replacements. It is important to explore the opportunities in Rendang District with the highest goat population in Karangasem Regency (BPS-Bali 2014), along with the policy of the Bali Regional Government who allow smallholder farmers to grow roughage in conservation forests, as well as providing them with Simantri (Integrated Farming System), Bansos (Social Aid) and LM3 (Independent Rooted Community Institution) Programs. Water is available throughout the year and goat rearing has significant synergistic interactions with growing vegetable crops. A database identifying constraints to, challenges of and opportunities for rearing goats in Bali Province has begun in Banjar Belulang, Sepang Village (Doloksaribu *et al.* 2014b) and in Karangasem Regency (Doloksaribu *et al.* 2014a). This study was conducted to observe growth performance and flock dynamics (birth, death, survival and sales) of kids under smallholder production systems in Karangasem Regency, Bali Province.

## Materials and Methods

There were 63 goat owning families who had a total of 1,163 goats in Pempatan Village, Rendang District in Karangasem Regency. These farmers were interviewed and data was recorded through observations, interviews, and focus group discussions during visits to farms. Karangasem Regency is situated 08°33'07" to 08°10'00" south and 115°23'22" to 115°42'37" east with ambient temperatures between 19 to 27.5 °C, and annual average relative humidity of 77%, rainfall of 1,428 mm and wind velocity of 9 knots ([www.bmkg.go.id](http://www.bmkg.go.id)).

A total of 1,265 sets of data (age; sex; body weight and length; chest circumference and depth; height at withers and rump) was collected on 499 kids from four months of observations (April, June, August and September). Age of kids were estimated from their dentition status ( $I_0$ ) while their birth dates were estimated by using the farmer's data and they were confirmed by looking at the condition and size of the dam's mammae or date of post-partum mating. Kids were weaned, on average, at about 135 days of age.

All 63 goat owning families completed questionnaires on a range of topics including goat feeding and breeding management, income from goats and other sources, as well as the demographics of people living on the farm. The relationships between liveweights and other factors were established using a linear regression analysis while correlations of the goats liveweight with other factors were computed by using SPSS version 22.

## Results and Discussion

### Kid's Births

In 2014, 568 kids were born from 413 kidding occurrences from 362 productive females that had  $I_0/I_1/I_2/I_3/I_4$  or toothless dentition and the ratio between female and male kids was 289:279, i.e. 1:1. Of the 413 kidding occurrences, 65% were single born (268 kids), followed by 33% twins and 2% for triplet and quadruplet born kids (Table 1). They were born from the 1<sup>st</sup> to 11<sup>th</sup> parity of does that gave about 100 kidding occurrences for both the first and second parity.

Table 1. Parity, gender and type of birth (singles, twins, triplets, quadruplets) of kids born in Karangasem Regency in 2014

Parity	Singles			Twins				Triplets			Quadruplets	Kidding occurrence	Kids born	Litter size
	F	M	Σ	F-F	M-M	F-M or M-F	Σ	F-F-M or M-M-F	M-M-M	Σ	F-F-M-M			
1 <sup>st</sup>	56	44	100	1	4	11	16	1	-	1	-	117	135	1.15
2 <sup>nd</sup>	34	39	73	10	6	12	28	-	-	-	-	101	129	1.28
3 <sup>rd</sup>	18	20	38	8	9	13	30	1	2	3	-	71	107	1.51
4 <sup>th</sup>	20	11	31	8	5	11	24	-	1	1	-	56	82	1.46
5 <sup>th</sup>	6	7	13	4	-	14	18	1	-	1	-	32	52	1.62
6 <sup>th</sup>	3	4	7	-	-	5	5	-	-	-	-	12	17	1.42
7 <sup>th</sup>	1	1	2	2	2	3	7	1	-	1	-	10	19	1.90
8 <sup>th</sup>	-	2	2	1	2	1	4	-	-	-	-	6	10	1.67
9 <sup>th</sup>	-	-	-	-	-	-	-	-	-	-	-	-	-	-
10 <sup>th</sup>	-	1	1	1	1	2	4	-	1	1	-	6	12	2.00
11 <sup>th</sup>	1	-	1	-	-	-	-	-	-	-	1	2	5	2.50
Total	139	129	268	35	29	72	136	4	4	8	1	413	568	1.65

Kidding occurrences for the 6<sup>th</sup> to 11<sup>th</sup> parity varied from 2 to 12. There was a tendency that litter size increased from 1.15 in the 1<sup>st</sup> parity to 2.5 in the 11<sup>th</sup> parity although for the 4<sup>th</sup>, 6<sup>th</sup> and 8<sup>th</sup> they declined and the average litter size was 1.65 kid's born per doe in 2014.

### Kid's Mortality and Survival Rates

From the total of 568 born kids in Karangasem Regency in 2014, only 8.6% died and 91.4% or 519 kids survived until they reached weaning age at about 4.5 months (Table 2). The ratio of kid's deaths between females and males was 22:27, i.e. 1:1.2. The highest rate of mortality was 63.3% for twin born

kids followed by 26.5% for single born kids and the rest was about 10% for triplet and quadruplet born kids. Kids mostly died during parturition or early after birth.

Table 2. Flock dynamics (birth, death, survival, sold and that were reared) of kids born in 2014 in Karangasem Regency

No of kid's	Singles			Twins			Triplets			Quadruplets			Total		
	F	M	Σ	F	M	Σ	F	M	Σ	F	M	Σ	F	M	Σ
Birth	139	129	268	142	130	272	6	18	24	2	2	4	289	279	568
Death	8	5	13	13	18	31	1	3	4	0	1	1	22	27	49
Survival	131	124	255	129	112	241	5	15	20	2	1	3	267	252	519
Sold	3	6	9	5	9	14	-	-	-	-	-	-	8	15	23
Were reared	128	118	246	124	103	227	5	15	20	2	1	3	259	237	496

When the percentage of kids that survived to those born, based on their type of birth was calculated, single born kids has the highest percentage, followed by twin, triplet and quadruplet born kids being 95%, 89%, 83% and 75%, respectively.

### Kid's Sales

Kids were usually sold for breeding stock unless as Mecaru kids, although they were also sold for both breeding stock and Mecaru ritual purposes. More male kids were sold (female : male 8:15, i.e. 1:1.87; Table 2). No triplets or quadruplets were sold. Mecaru kids aged 1 month up to 4.5 months of age were popular in Karangasem Regency as their prices were cheaper than older Mecaru goats. As long as a kid had all black coat colour, regardless of its body weight or age, it officially can be used as a sacred animal in Mecaru rituals. Mecaru kids were sold for Mecaru rituals or breeding stock for Mecaru goat farms. Farmers tended to breed more black goats as there were more profits from selling Mecaru goats, particularly the pre-weaned kids. The total value for 3 Mecaru, 6 female and 14 male breeding animals sold was IDR 35 M (Table 3).

Table 3. Prices of goats sold in Karangasem Regency

Description	Age (months)	Prices (IDR)	Description	Age (months)	Prices (IDR)
Mecaru	3-12	1,000,000- 2,000,000	Meat goats	10-18	750,000- 1,500,000
Male breeding stock	6-10	1,500,000- 1,800,000	Culled males	>36	750,000- 1,500,000
Female breeding stock	6-10	1,250,000- 1,500,000	Culled females	>36	750,000- 1,500,000
Male Idul Qurban	12-24	1,800,000- 2,500,000			

### Biological Measurements of Kids

Growth performance of goats can be measured and expressed as body weight at various ages. The growth of kids prior to weaning is critical to their economic value, as birth and weaning weights are related to kid survival and postnatal development (Luo *et al.* 2000), and minimizing the cost of rearing kids provides more profits to farmers (Malik *et al.* 1986).

Data on the birth weight of kids in this study was relatively small (60 animals) compared to the total dataset, therefore birth weight and body dimensions analysis were limited to the kids aged 1 to 7 days for more accurate results. The average birth weight of kids in this study (3.1 kg) was relatively high, probably as almost half of the kids born in 2014 were single born kids that are generally heavier than twin or triplet born kids (Tables 1 and 2). It appears that newly born kids received sufficient quantity and quality of colostrum and milk from their dams, as the average weight and daily gain of kids at the end of the first week was 4.3 kg and 171 g/d, respectively. There were no significant differences ( $P>0.05$ ) in the average liveweights at weaning, to 135 days of age, between females ( $9.8 \pm 0.2$  kg,  $n=432$ ) and males ( $10.5 \pm 0.3$  kg,  $n=404$ ) although males tended to be heavier. However, the average liveweight gain of males ( $143 \pm 5$  g,  $n=197$ ) were significantly higher ( $P<0.01$ ) than that of the females ( $113 \pm 4$  g,  $n=207$ ).

The liveweights and average daily liveweight gain of kids in this study were comparable to Etawah kids of the same ages, reared in Java (Sodiq 2012) and Anglo Nubian crossbreds reared in the Indonesian Research Institute for Animal Production in Bogor (Praharani 2014), although the liveweights of kids at

60, 90 and 135 days of age in this study were slightly higher compared to Anglo Nubian crossbred kids (Praharani 2014) but lower than the Etawah kids reared in Java (Sodiq 2012). The liveweights and average daily liveweight gains of kids after the first month in this study tended to be lower than reported by Sodiq (2012) as the kids were not fed any concentrates or other feeds to stimulate their rumen development. The kids in this study probably ate the roughage or dagdag provided for mature goats and it is presumed that these kind of feeds or dam's milk had not met the nutrients required by kids for their optimal growth after their first month. The average liveweights of a total of 499 female and male kids were 4.3 kg at seven days of age, 7.4 kg at 30 days, 10.5 kg at 60 days, 13.5 kg at 90 days and 18.2 kg at 135 days of age.

Liveweights of kids born in Karangasem Regency in 2014 had strong positive significant ( $P < 0.01$ ) linear correlations with age (0.789), ADG (0.233), chest circumference (0.906), chest depth (0.900), body length (0.869), height of withers (0.854) and rump height (0.853), but had strong negative significant ( $P < 0.01$ ) linear correlations with kidding season (-0.537) and FAMACHA<sup>®</sup> scores (-0.275). Housing management i.e. battery and colony systems as well as the gender of the kids did not significantly affect their liveweight ( $P > 0.05$ ).

## Conclusion

Productivity of kids under smallholder production system in Pempatan Village, Karangasem Regency was profitable and can contribute significantly to the improvement of productivity of goat farms as growth performance was relatively high, and low FAMACHA<sup>®</sup> score, low mortality but high prices for selling breeding stocks particularly for the Mecaru kids. Furthermore, rearing kids in Pempatan Village, Karangasem has been supported by low initial investment, low production costs, minimal or no additional expenditure in feeds and low dependence on external inputs as well as Bali Regional Government's Programs.

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# Daily Activities and Propolis Production of *Trigona* Bee Keeping in Three Nest Types

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## Abstract

This research was aimed to assess bee activity including going out and into the nest and propolis production of *Trigona* bee kept in various nest types. A completely randomized design with three treatments and fifteen replications was applied. Three types of nests made of wooden box, large bamboo and small bamboo were used for bee keeping. Data were analyzed using analysis of variance. Calculation of bee activities including going out and into the nest was done every morning and afternoon for ten minutes for each nest type. Two kinds of propolis such as honey and pollen wrapping propolis and nest wall propolis were weighed. The result showed that the activities of bee kept in wooden box was the highest with average of  $80 \pm 4$  times significantly different from large and small bamboo with average  $63 \pm 3$  and  $62 \pm 3$  times, but activities in large and small bamboo were not significantly different ( $p < 0.05$ ). The weight of propolis in the box was higher ( $43.50 \pm 18.9$  g) than average weight of honey + pollen wrapper. Propolis on the wall with the average weight  $17.00 \pm 6.02$  g was significantly different with propolis in large bamboo with honey + pollen wrapper ( $20.50 \pm 11.4$  g). Propolis on the wall with the average weight of  $9.33 \pm 4.9$  g and propolis in the small bamboo has honey + pollen wrapper with the average  $23.00 \pm 13.5$  g. Propolis on the wall had average weight of  $8.47 \pm 2.17$  g was significantly ( $p < 0.01$ ) lower than propolis on the other nest types. Propolis wraps for honey + pollen for all various nest types is significantly different with propolis on the walls ( $p < 0.01$ ). Based on these result it could be concluded that the highest bee activities including activity was expressed by *Trigona* bees kept in the wooden nest (box).

**Keywords:** nest, propolis, *Trigona*

## Introduction

*Trigona* bee farming have been carried out by rural communities as an alternative business a good prospect for development because it produce products of high economic values including honey, wax and propolis. In their natural habitat, *Trigona* bees are always found in trees, wood and bamboo, they preferred stable condition and trying to be away from predators. The main activities included "in and out of the stumps, maintaining predator attack, removing dirt in the nest, and foraging (Sihombing, 2005). Going out and back into the nest was influenced by the availability of food, temperature and humidity (Hilario *et al.*, 2003). The frequency of activity was increased when there was an abundance of food resources surrounding the hives. Beside for consumption and health, propolis can be used for nest protection and sterilizing the hive from pests such as bacteria, fungi and viruses. According to Hasan (2010), the propolis contained antibiotics for curing some human diseases, as it had high antioxidant and vitamin C, as flavanoid. The levels of flavonoids propolis *Trigona* reached 4%, while propolis *Apis cerana*, *mellifera* and *dorsata* 1.5%.

Bee farming practices by farmers, was generally using different types and size of stumps/nests according to the local available materials or local culture. This affected the activity of bees to be in and out during the day, and hence, propolis production. Therefore, it is necessary to examine appropriate types and forms of cages or stumps used for keeping *Trigona* bees, and measure the propolis production. The main objective of this study was to evaluate and measure the activity "in and out" of the nests and propolis production from *Trigona* bee using different types of nests.

## Materials and Methods

The material used in this study is *Trigona* bees placed on 3 different nests. The tools used are digital scales (brand Shuma) capacity of 3000 gram with a sensitivity of 1 gram to weigh *Trigona* bee propolis and counter check to calculate the activity of bees *Trigona*, thermo hygrometer for measuring the temperature

and humidity of the environment around the sites. Materials used were made of wood *borok* with a thickness of 2 cm and the size of 35 cm x 17.5 cm x 10 cm, bamboo with a diameter of ± 6 cm and bamboo with diameter of ± 8 cm. A completely randomized design was applied in this study, with 3 treatments including 3 types of nests (1) wooden box; 2) small bamboo; and 3) large bamboo), each treatment contained 15 units as replication, with the total numbers of nest of 45 units.

The measurement of *Trigona bea* activity was done in the morning and afternoon on the daily basis, using the Counter Check by standing in front of a nesting colony for 10 minutes. Propolis produced was weighed at the end of the study, collected from wrapping honey, pollen and propolis scattered on the nest walls.

### Data Analysis

Data obtained was analysed using Analyses of Variance (Anova) to see the difference between treatments, and tested further using Duncan test (Steel and Torrie, 1993).

## Results and Discussion

### Activities Going Out and Into The Nest *Trigona Beae*

The frequency of going out and into the nest of *Trigona bea*, significantly affected the production of propolis. The higher the frequency, the higher the propolis production was obtained (Table 1 and Fig. 1).

Table 1. The average frequency of *Trigona bea* going out and into the nest

Week	Nest Types		
	Box	Large bamboo	Small bamboo
1	85 ± 9 <sup>a</sup>	61 ± 5 <sup>b</sup>	65 ± 3 <sup>b</sup>
2	82 ± 17 <sup>a</sup>	66 ± 8 <sup>b</sup>	63 ± 4 <sup>b</sup>
3	77 ± 14 <sup>ab</sup>	65 ± 8 <sup>b</sup>	61 ± 5 <sup>b</sup>
4	77 ± 14 <sup>a</sup>	60 ± 6 <sup>b</sup>	56 ± 3 <sup>b</sup>
5	75 ± 15 <sup>a</sup>	59 ± 4 <sup>b</sup>	64 ± 4 <sup>b</sup>
6	76 ± 14 <sup>a</sup>	61 ± 3 <sup>b</sup>	61 ± 3 <sup>b</sup>
7	84 ± 12 <sup>a</sup>	65 ± 3 <sup>b</sup>	63 ± 5 <sup>b</sup>
8	82 ± 11 <sup>a</sup>	68 ± 4 <sup>b</sup>	61 ± 3 <sup>b</sup>
Average	80 ± 4 <sup>a</sup>	63 ± 3 <sup>b</sup>	62 ± 3 <sup>b</sup>

Description : Different superscripts in the same row and column indicate significant differences (P < 0.05).

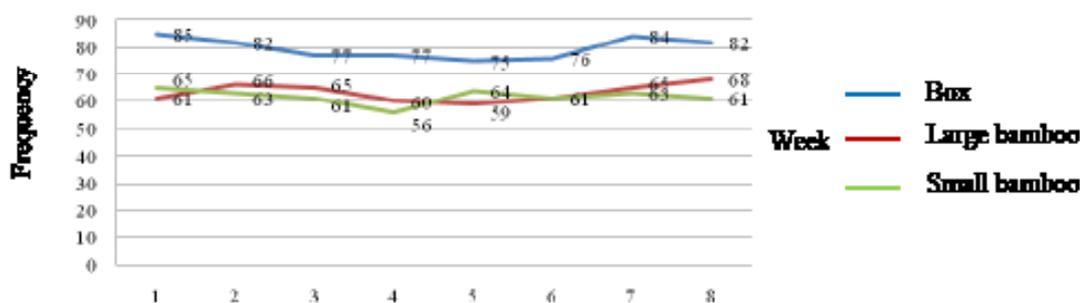


Figure 1. Frequency of *Trigona bea* going out and into the nest based on nest types

The results showed that the activity going out and into the nest *Trigona bea* on the type nesting of the box (80±4 times) were significantly different (P < 0.05) compared with large bamboo (63±3times) and a small bamboo (62±3times). This figures was higher than the results of Hermawan (2007) of 20.89±6.64times. The higher activity was probably affected by environmental conditions of the location with the average temperature of 26°C and humidity of 64%, which was suitable for *Trigona bea* farming. According to Erwan (2003), many bee worker went out for feed hunting such as nectar from plants and flowers in the morning, because the volume of nectars were still available, and in sufficient volumes as those were accumulated from thesecretion that occurred since the afternoon until evening. Whereas, during the daytime, most of the bee workers went out for drinking waters.

## Propolis Production

Propolis is the most main products produced by Trigona bees. Some other benefits of propolis, including protecting bee colonies from diseases (Salatino et al., 2005). Propolis production of three types of nests can be seen in Table 2.

Table 2. The average of propolis production from different types of nests

Propolis	Nest Types		
	Box (gr)	Large bamboo (gr)	Small bamboo (gr)
Wrapping Honey and Pollen	43,50±18,9 <sup>a</sup>	20,50±11,4 <sup>b</sup>	23,00±13,5 <sup>b</sup>
Wall	17,00±6,02 <sup>c</sup>	9,33±4,9 <sup>d</sup>	8,47±2,17 <sup>d</sup>

Description: different superscripts in the same row and column indicate significant differences ( $P < 0.01$ )

The results showed that the propolis production of wraps honey and pollen (43.50±18.9 g) compared to Trigona bee propolis on a wall made of wooden nesting (box) was higher ( $P < 0.01$ ) compared with large bamboo (20.50 ±11.4 g) and a small bamboo (23.00 ±13.5).

## Temperature and Humidity

Temperature and humidity in the study fairly stable with an average temperature of 26 °C and a humidity of 64%. Stable temperature and humidity affected bee activities and influenced the by the types of flowers for bee feed in the form of nectar and pollen. Sedgley (1991) suggested that high activity would affect the propolis production of Trigona bee. Environmental weather conditions greatly affected the activity of bees, among others, temperature, light intensity and humidity and wind speed.

## Distance Sources of Feed with Nesting

Source sufficient feed bee Trigona affect production in the form of propolis, honey, pollen and the number of tillers. Food that is available around the site of research has a diversity ranging from palm, mango, coconut and various other crops. Distance sources of feed with nesting ± 5 meters, it is easier for bees to feed.

The distance between the bee colonies, with the sources of food had a big influence on honey and propolis production. This is evident from the production of honey, pollen, propolis and the number of tillers. Sarwono (2001) states that all species of flowering plants (plants of forest, agricultural crops, plantation crops, horticultural crops and wild plants) which contains elements of nectar as an ingredient of honey, pollen, and propolis can be used as feed for bees. Free (1982) states that the bee can visit several hundred flowers to collect nectar or pollen that much as a source of feed. The distance for Trigona bees to find food sources, was stated within a radius of approximately 500 meters (Baconawa, 1999).

## Conclusion

The frequency of Trigona bee, went out and into the nest, in the different types of nests of wooden box was higher than those kept in large bamboo and small bamboo, which was correlated to the higher propolis production. Propolis were mostly found in the wrapping of honey and pollen in the box as compared to those on the wall of nests.

## Acknowledgement

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# Harmony between Livestock Behaviors: Birth Time and Sites Selection Behaviors in Sheep and Goats

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## Abstract

*Study of sheep and goat's peri-parturition behaviors is very important to understand proper handling management of sheep to get good performances of both mothers and offspring. Behaviors of sheep and goats in selecting birth sites, birth time were investigated and were discussed in relation to their natural behaviors of maternal-new born bonding pattern. Sixty nine (69) heads of Booroola Merino ewes that were in late pregnancy in May-June 1991 and eighty two (82) heads of Angora goats were observed their birth time and birth sites. Birth times were grouped into two day time period (06.00-18.00) and night time (18.00-06.00), and birth sites were categorized as protected and open areas. Data were analyzed with Chi-square ( $\chi^2$ ). The results show that birth time in sheep was not significantly different between day and night time period. In contrast, goats gave birth mostly in day time period. In selecting birth sites, sheep chose both protected areas and open sites in similar preferences, while goats chose more protected areas. The findings are in harmony in sheep as they have close contact bonding pattern. Similarly, with the lying-out maternal newborn kids bonding pattern in goats, more births occurred during daytime period and more in protected areas, are reasonable for protecting possible mis-mothering cases. It is concluded that selection of birth time and birth sites occurred in harmony with bonding pattern either in sheep or goats. The harmony is important to animal welfare concerns for sustainable animal production.*

*Keywords: Birth time, birth sites behaviors, goats, harmony, sheep,*

## Introduction

Fecund breeds of sheep or goats often produce more than two lambs or kids. Loss of newborn animals is greater in multibirths than in single births, due to the possibility of mismothering (Alexander, 1984). It is important to have management procedures which ensure the maximum survival of the litter. It is important to understand the parturition behaviour and the characteristics of the paddock in which parturition occurs.

The time of birth is an important consideration in relation to parturition behavior. This aspect has been documented by previous work (Yamin, 1991, Ayhan *et al.* 2012). This information can be useful as basic data for conducting further behavioral studies or as a basis for developing intensive periparturient management strategies with these species.

The effect of physical environmental features in the paddock, on the choice of birth site is also of interest. Such features could include open areas, shady areas, area close to vertical objects (fences, shed etc), feeding or drinking spots, elevated and sloping areas, as all of these features could be seen to provide more or less safe places for newborn lambs or kids.

This experiment aims to:

1. study birth time pattern behaviour of the merino ewes and angora goats
2. study birth sites choice behavior of the merino ewes and angora goats
3. analyze the harmony of the birth time and sites pattern with the maternal-offspring bonding pattern in sheep and goats.

## Materials and Methods

### Materials

The animals used in this experiment were female sheep and goats having age of more than 2 years old and they were included: a). Sixty nine (69) heads of Booroola Merino ewes that were in late pregnancy, b). Eighty two (82) heads of Angora goats that were in late pregnancy.

All paddocks were located in the sheep and goat section, The University of Queensland, Gatton College, Lawes, Queensland Australia. The paddock used for sheep had 2.4 ha (Bates 5A) and for goats it was 2.1 ha (Bates 4B and 4C). A binocular was used to observe experimental sheep and goats for specific births times and sites and other general situations relating to parturition.

### Procedures

To study birth time distribution, the observations were conducted from 06.00 until 18.00 h each day. The times of birth were recorded in three hour intervals during that period. The period between 18.00-06.00 h was considered as one period due to the limitation of time and labour to conduct observations. The complete division of each 24 hour period was 18.00-06.00 h, 06.00-09.00 h, 12.00-15.00 h, and 15.00-18.00 h.

To study the effect of paddock features on birth site distribution, the paddock was classified into shady areas, areas close to vertical objects and open areas. Shady areas were defined as substantial areas of shade such as under trees, sheds and the wall in paddocks. Areas close to vertical objects were defined as non-shaded areas close to (within 1 meter) of any vertical object which included fences, and feeding or drinking troughs. Open areas were any other exposed areas beside the previous two areas. The partitioning of the paddock into 3 types of features resulted into approximately 363 m<sup>2</sup> of shady areas, 441 m<sup>2</sup> of areas close to vertical objects and 866 m<sup>2</sup> of open areas. To analyze the birth site distribution associated with the similar size of natural features, shady areas and area close to vertical objects were combined resulting in 804 m<sup>2</sup> of protected area and 866 m<sup>2</sup> of open area.

The observations were conducted every two hours from 06.00 to 18.00 h each day. The birth sites used during the night were determined by the presence of characteristics associated with birth such as after-birth (placenta), blood and any birth fluids. Where birth sites could not be determined, no recordings were made.

### Data Analysis

Chi-Square ( $\chi^2$ ) Goodness-of-fit test (Siegel and Castellan, 1988) was used to analyze the distribution of birth periods. The mathematical model used to test the null hypothesis ( $H_0$ ) is:

$$\chi^2 = \sum_{i=1}^k \frac{(O_i - E_i)^2}{E_i} \text{ with } df = k - 1$$

where:  $O_i$  = the observed number of cases in the i-th category  
 $E_i$  = the expected number of cases in the i-th category when  $H_0$  is true  
 $K$  = the number of categories

## Results and Discussions

### The Distribution of Time of Birth in Sheep and Goats

Figure 1a shows that in sheep the frequencies of births occurring between time periods during day time were not significantly different (20.3%, 13%, 13% and 7.3%, respectively;  $X^2=4.40$ ;  $df=3$ ;  $p>0.05$ ). However in goats the percentage of births between 06.00-09.00 h (7.1%) was significantly lower than in the other day time periods with 22.6%; 31.0% and 17.9% of births respectively for intervals between 09.00-12.00 h; 12.00-15.00 h and 15.00-18.00 h. There were no significant difference in percentage of births between the last three day time periods ( $X^2=2.1$ ;  $df=2$ ,  $p>0.05$ )(Figure 1a). By grouping the 4 intervals of time during day light into one period (06.00-18.00 h), it accounted for 53.6% of birth frequency. This was not significantly different from the frequency of birth (46.4%) occurring overnight (18.00-06.00 h) ( $X^2=0.36$ ;  $df=1$ ;  $p>0.05$ ) (Figure 1b). In contrast, for the goats, the analysis showed that there were significant differences between the two intervals of day light and overnight (21.4% and 78.6%, respectively;  $X^2=27.43$ ;  $df=1$ ;  $p<0.001$ ) (Figure 1b).

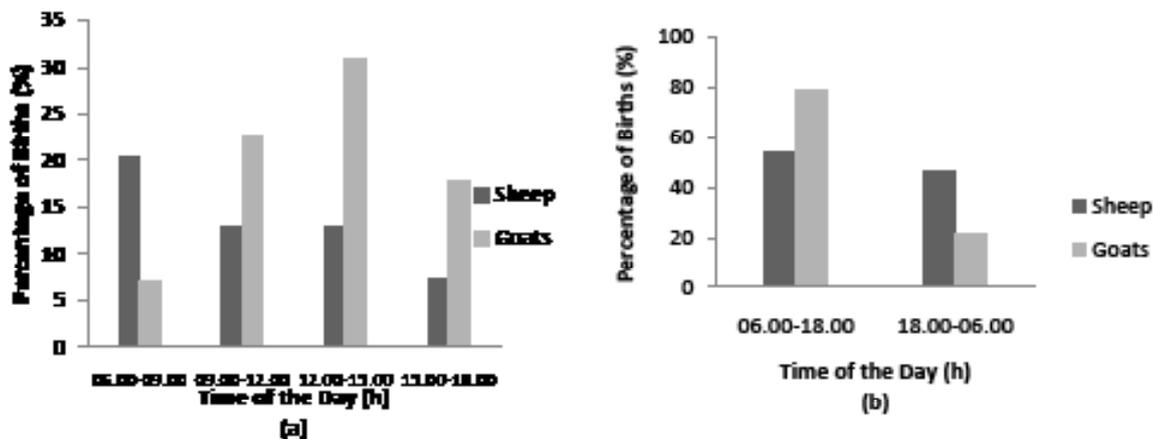


Figure 1. Distribution of time of birth in sheep and goats during the day time (a) and between day and night time (b)

### Birth Site Distribution in Sheep and Goats

By classifying the features of the paddock into three types of birth types, it was found that 44.1% of births occurred in open areas, 32.4% in the shady areas and 23.5% were close to vertical objects (Figure 2a) and it was not significantly different between the three areas ( $X^2=4.35$ ;  $df=2$ ;  $p>0.05$ ). However the does preferred to give birth in shady areas more than areas close to vertical objects (47.6% vs 23.2%, respectively,  $p<0.01$ ). No significant different between open areas and vertical objects ( $p>0.05$ ). When shady areas and vertical objects were combined to become protected areas, it shows that in ewes, births occurring associated with protected or open sites were not significant different (55.9% vs 44.1%;  $X^2=0.94$ ,  $df=1$ ,  $p>0.05$ ). However, the does chose more protected areas than in open areas ( $X^2=14.1$ ;  $df=1$ ;  $p<0.01$ ) (Figure 2b).

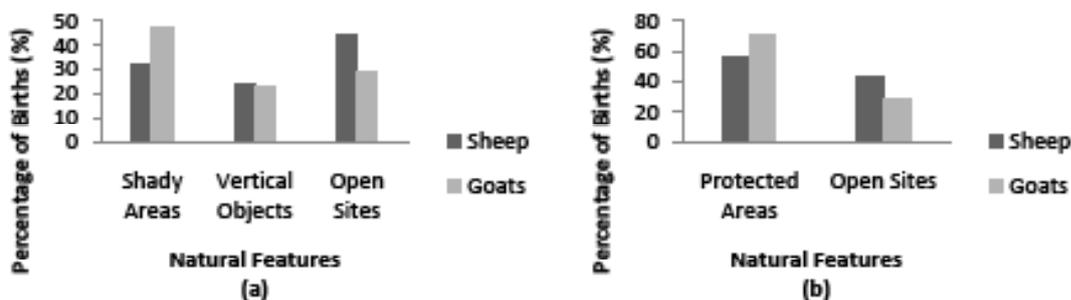


Figure 2. Birth site distribution in sheep and goats at 3 different sites (a) and 2 different sites (b)

### Harmony between Behaviors: Birth Time, Birth Site and Maternal-Offspring

The different pattern of births between sheep and goats was interesting observation. One basis for the difference in the diurnal pattern of parturition between goats and sheep may be related to basic behavioral differences that exist between those species. Lambs are categorized as follower types and kids as stayer types (Lickliter, 1984; Pascal *et al*, 2007; Hernandez *et al* 2012). This could be a basic factor influencing the time and site of births. Time of birth may not be so important for newborn lambs, as they always follow their dams. Therefore, it may be less risky for sheep to give birth during the night or early morning since close contact is maintained with the young at all times and the risk of predation may be lessened. Similarly, type of hiding behavior of young kids (characterized as stayer type animals) may have encouraged the development of parturition during day light hours as a means of allowing the selection of better hiding sites and protection from predators and mismothering problems.

### Conclusion

Harmony between parturition behaviors in sheep and goats could have contributed to better animal handling management, animal welfare and to the preservation of these species for sustainable animal industry.

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**THEME B.**  
**FEED TECHNOLOGY**

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# Fermentation Kinetics of Palm Oil Plantation by-Product Based Diet

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## Abstract

Plantation by-product is one of many alternative feed sources for ruminant. Nowadays, palm oil tree leaves is the most potential plantation by-product used as source of fibrous feed. General characteristic of the feed source is high level of fiber content and low digestibility. Therefore addition of high quality diet such as concentrate is required. A study was undertaken to investigate the degradability and fermentation kinetics of complete diet consisted of palm oil leaves and concentrate. Three complete diets were investigated by using *in sacco* and *in vitro* method to determine the degradation rate in the rumen as well as fermentation kinetic in the rumen system, by using complete randomized design. Feed A as control feed consisted of King grass 60%+Gliricidia 40%; Feed B consisted of King grass 30%+concentrate 70%; and Feed C consisted of palm oil leaves 30%+concentrate 70%. The study measured dry matter and organic matter degradability and profile of methane gas, NH<sub>3</sub> and VFA produced during the fermentation of feed in the rumen system. Results showed that degradation rate of Feed C (34.71%) was lower than that of Feed B (36.47%) and Feed A (27.40%) ( $P < 0.05$ ). The organic matter degradability of Feed A (25.96%) was the lowest among the three Feeds ( $P < 0.05$ ). The concentration of ammonia and VFA in Feed C was the highest among the three Feed treatment ( $P < 0.05$ ). The Feed C produced more gas total and methane gas during the rumen fermentation by rumen microbes, compared to those the two Feeds. The conclusion is that that complete feed based on King grass and Gliricidia leaves has lower degradation rate in the rumen. Complete feed based on palm oil leaves supplemented with concentrate produced more VFA and gas methane during the fermentation process in the rumen.

**Keywords:** degradability, fermentation kinetics, methane, palm oil plantation by-product

## Introduction

Palm-oil leaves is potential feed sources for ruminant animals in term of availability and nutrient content. Each acre of palm-oil plantation provides about 0.66 ton/year of leaves that can be used as source of fiber for ruminant feed (Diwyanto, *et al.*, 2003). The leaves contains 21.52% crude fibre, 14.12% of crude protein and 4.46 MJ/kg gross energy (Elizabeth and Ginting, 2003). According to Mathius *et al.*, (2004), fresh leaves of palm-oil tree can substitute fress grass up to 30% in complete feed, and can be increased up to 50%, when the leaves was ensilaged (Wan Zahari *et al.*, 2003). The limiting factor for palm-oil leaves utilisation for ruminant feed is the high lignin content (13.79% - 20%) (Badan Penelitian dan Pengembangan Pertanian, 2012). To be useful as ruminant feed, beside the chemical value, the palm-oil leaves must also has biological value. The biological value can be determine by using *in sacco* and *in vitro* techniques. The aim of experiment was to investigate the degradability and fermentation kinetics of complete diet consisted of palm oil leaves as basal diet.

## Materials and Methods

Experiment tested three complete feeds by using *in sacco* and *in vitro* techniques. Fresh King grass (*Pennisetum purpupoides*) and commercial feed (concentrate) were used as common basal diet for ruminant to test the palm-oil leaves basal diet. Gamal (*Gliricidia sepium*) leaves was used to substitute concentrate as commercial feed supplement. The composition and nutrient content of each complete feed are presented in Table 1 and Table 2.

Table 1. Composition of three complete feeds used in the experiment in dry matter (DM) based

	Feed A	Feed B	Feed C
King grass (%DM)	60	30	0
Palm-oil leaves (%DM)	0	0	30
Gliricidia leaves (%DM)	40	0	0
Concentrate (%DM)	0	70	70

Table 2. Nutrients content of three complete feeds used in the experiment

Complete feed	Crude Protein (%)	Fat (%)	Crude Fibre (%)	Ash (%)	Energy (%)
Feed A	14.98	2.83	38.54	10.07	4325
Feed B	10.58	7.52	28.13	9.94	3940
Feed C	9.19	7.76	32.73	7.82	4001

Two rumen-fistulated cattle were used to test DM and OM degradation of three complete feed in the rumen. The animals were fed by king grass (*Pennisetum purpureoides*) and concentrate. The DM and OM degradation was examined by using Nylon bag (pore size of 45  $\mu$ m; 6 cm x 12 cm in size), and prepared according to the method described by Orskov *et al.* (1980). The sample size of 5 g was recommended by Kempton (1980) for the determination of the degradation of feeds. Two sets of bag were inserted into rumen of two rumen-fistulated cattles with the incubation times of 48 hours. Each samples has 6 replicates (2 animals x 3 series of 48 h incubation). After withdrawal from the rumen, each bag was washed then dried in an oven at 60 °C to constant weight following procedure of Kempton (1980) for DM and OM analyses.

Profile of methane gas, concentration of NH<sub>3</sub> and VFA produced during the fermentation of feed in the rumen system were determined by using *in vitro* technique of Theodorou and Brooks (1990); Theodorou *et al.*, (1994). Feed samples, buffer solution and rumen fluid were mixed and placed in serum bottle (150 ml in volume) and flushed by using CO<sub>2</sub> to make anaerob condition in the bottle, then incubated in waterbath 39 °C for 48 hours. Total and methane gas produced were measured by using glass syringes at 3, 6, 9, 12, 18, 24, 30, 36, 42, 48 h of incubation. Sample for NH<sub>3</sub> and VFA concentration analysis were taken after 48 hours of incubation. The NH<sub>3</sub> concentration of the medium was determined using the Conway technique developed by Conway and O'Malley (1942). The VFA concentration was determined using Gas Liquid Chromatography (GLC; Hewlett Packard, 3700, USA). All data was analyzed using IBM SPSS statistics ver. 20 following the complete randomized design (Steel and Torrie, 1980)

## Results and Discussion

Dry matter and organic matter degradation of three complete feeds in the rumen during 48 hours of incubation are showed in Table3.

Table 3. The DM and OM of three complete feed degraded in the rumen during 48 hours

	DM degraded (%)	OM degraded (%)
Feed A	27.40 $\pm$ 0.736 <sup>a</sup>	25.96 $\pm$ 0.774 <sup>a</sup>
Feed B	36.47 $\pm$ 1.263 <sup>c</sup>	37.62 $\pm$ 1.664 <sup>b</sup>
Feed C	34.71 $\pm$ 1.953 <sup>b</sup>	36.85 $\pm$ 2.737 <sup>b</sup>

The same superscript in the same coloum indicated not significant different (P>0.05)

The amount of DM Feed B degraded in the rumen was significantly higher than those Feed A and Feed C (P<0.05). The amount of DM Feed A degraded after 48 hours of incubation time was the lowest among the three Feed examined (P<0.05). Similar pattern also found in the amount of OM degraded in the rumen after 48 hours. Differences in the crude fibre content of the three complete feed was the most reason for the differences in degradability. Feed B contain the lowest crude fibre compared to the other two Feed, while Feed A contain the highest crude fibre (Table 2) resulting in lowest amount of DM and OM degraded in the rumen (Table 3).

The components of Feed B are grass and concentrate, while Feed A is grass and Gliricidia leaves and Feed C is palm-oil leaves and concentrate. It indicated that concentrate is more degraded than Gliricidia

leaves, since Gliricidia leaves contains more crude fibre than that of concentrate. While Palm-oil leaves contains more lignin than those of grass leaves (Badan Penelitian dan Pengembangan Pertanian, 2012). Lignin is the nutrient that hardly degraded by rumen microbes. The end products of rumen fermentation of three complete feed examined are presented in Table 4.

Table 4. Gas produced, NH<sub>3</sub> and VFA concentration of rumen fermentation of three complete feeds during 48 hours of incubation

	Feed A	Feed B	Feed C
Gas total (ml)	108.67±16.23 <sup>a</sup>	112.67±21.54 <sup>a</sup>	161.50±29.61 <sup>b</sup>
Gas CH <sub>4</sub> (ml)	38.83± 5.31 <sup>a</sup>	38.33± 4.23 <sup>a</sup>	53.0± 3.85 <sup>b</sup>
Gas CH <sub>4</sub> /Gas Total (%)	39.72± 6.05 <sup>b</sup>	34.5± 3.27 <sup>a</sup>	33.44± 4.38 <sup>a</sup>
NH <sub>3</sub> (mM)	13.85 ±0.72 <sup>a</sup>	17.39 ±3.35 <sup>a</sup>	24.49 ±1.71 <sup>b</sup>
VFA Total (mM)	63.34 ±3.47 <sup>a</sup>	79.09 ±2.89 <sup>b</sup>	82.45 ±3.67 <sup>b</sup>

The same superscript in the same row indicated not significant different (P>0.05)

The profile of total gas and gas methane produced indicated that Feed C significantly produced more gas during the fermentation process in the rumen (P<0.05). While, total gas produced during the fermentation of Feed A and Feed B were relatively similar. However, when the gas methane was compared to total gas produced, presented as percentage of methane per total gas produced, indicated that proportion of gas methane per total gas produce from Feed A was higher than those of two other Feeds (Table 4). The only reason for this result is due to the highest crude fibre content of Feed A compared to Feed B and Feed C (Table 2). Feed A consisted of grass with Gliricidia leaves as feed supplement that contain more crude fiber compared to those Feed B and C which were supplemented with concentrate. Concentrate is high quality feed supplement that usually contain high energy and protein but low in crude fibre.

There was similar pattern in concentration of NH<sub>3</sub> and VFA recorded. Ammonia concentration indicated the amount of dietary protein degraded in the rumen. The concentration of NH<sub>3</sub> was not positively linear with the amount of protein contained in the Feed. Feed A contained the highest protein but recorded produce the lowest NH<sub>3</sub>. Supplementation of Gliricidia leaves might be the best reason for lower NH<sub>3</sub> concentration in Feed A. Most of leguminous leaves contain tannin that can inhibit protein degradation by rumen microbes. Concentration of VFA was positively linear with the OM contained in the Feed (Table 2) and OM degradation in the rumen (Table 3). Increasing in VFA concentration was followed by increasing in the amount of OM content and degraded in the rumen.

## Conclusion

It can be concluded that complete feed based on King grass and Gliricidia leaves has lower degradation rate in the rumen. Complete feed based on palm oil leaves supplemented with concentrate produced more VFA and gas methane during the fermentation process in the rumen.

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# Potential of Papaya (*Carica Papaya L.*) Leaf Flour in Animal Feed to Increase The Weight and Decrease The Ammonia On Broiler Excreta

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## Abstract

This study was conducted to determine potential of Papaya (*Carica papaya L.*) in Animal Feed to Increase the Weight and Decrease the Ammonia on Broiler excreta. The experiment used completely randomized design. Twenty four broiler chicks were allocated for four treatments, which is P0 (0% Papaya), P1 (5% Papaya), P2 (7% Papaya), and P3 (9% Papaya). Treatment started from day 21 to day 34. The results showed that Papaya 5% could reduce NH<sub>3</sub> slightly. In addition, the provision of 5% Papaya showed gain significant higher than control.

Keywords: ammonia, papaya leaf flour (*Carica papaya L.*), weight.

## Introduction

Chicken farm development in Indonesia has increased very amazingly, especially in the area of East Java which has developed very rapidly. Livestock Statistics data (2011), the total number of livestock production in East Java is 229.488.628 heads/year, 149.552.720 is broiler chickens, so broiler chickens accounted 65,1678% of the total livestock production in East Java. This figure is the largest contributor of chickens in Indonesia. Availability of broiler chicken population were used to meet the needs of consumers in East Java and the surrounding areas.

Chicken is homeothermic (warm-blooded), the body temperature of the chicken should retain for life and producing efficiently. Chicken trying to keep the body temperature remains in the range of 40.6 to 41.7 °C. Chickens will prevent their temperature rising by slowing the rate of metabolism and reduce feed consumption. Ambient temperature between 18-21 °C is a suitable temperature (thermoneutral zone) for optimal growth of chickens (North and Bell, 1990).

Chicken in tropic areas experiencing stress caused by high ambient temperatures. The average temperature ranges in tropics areas is between 20,63-33,30°C, with average ranges 26,81°C (BPS, 2004). Fluctuating ambient temperatures cause stress in chickens increased, immune system decreased, susceptibility to diseases, so the chicken productivity declined. The impact of high temperature is chicken consuming water more to reduce the heat stress. Excreta become more fluid and cause odor pollution. Efforts to control the smell of chicken excreta with zeolite, lime, and microbes have been tried. That such materials can reduce the production of NH<sub>3</sub> and sulfide. Another alternative to overcome the condition is by providing natural ingredients of papaya leaf powder in animal feed.

Based on phytochemical screening which has been done, the class of active compounds identified in the leaf papaya include *hydroquinone phenols, tannins, alkaloids, steroids and volatile oil* (Ardiansyah et al., 2002). Belutas extract efficacious cure diarrhea, dysentery drug, and can also cope albuminuria, the production of albumin in the urine. Such 5% Papaya leaf essential oil can inhibit the growth of *Staphylococcus bacteria aurius*, while 20% can inhibit bacterial growth *Escherichia coli*.

Negative impacts of farm is associated with livestock waste, in the form of production of excreta. The waste cause odor and gas. The odor of which come from the elements nitrogen. These elements will form NH<sub>3</sub>, nitrate, nitrite gas during the decomposition process. Air contains NH<sub>3</sub> can cause health problems for livestock and people surrounding the farm. Excreta odor problem is a problem that until now complained by breeder chickens because they often get protests from the local community. In addition to the influence of NH<sub>3</sub> on poultry have been reported, which can reduce the average growth, can reduce feed efficiency, respiratory tract damage (Cronic Respiratory Disease) and increases the activity of ND virus (New Castle Disease).

So there is a rationale for examining the effect of adding the papaya leaf flour to fodder for broiler excreta. Giving papaya leaf flour in the feed should be applied in broiler chickens in the tropics. The use of papaya essential oils in animal feed is one alternative to the negative impacts of odor management efforts in hopes of reducing the formation of the liquid excreta of broiler chickens. Based on this testing should

be further conducted to determine the level of provision of papaya leaf flour toward to NH<sub>3</sub> production of broiler excreta.

## Methods

This study is a laboratory experimental study using several methods such as spectrophotometric method with Nessler reagent for examination levels NH<sub>3</sub> (ammonia). Tools and materials used in this study is the Cobb strain broiler production strain CP 707 PT. Charoend Pokphand Jaya Farm as much as 24 head DOC (Day Old Chick) until the age of 37 days. As well as materials used in the examination of ammonia concentration (NH<sub>3</sub>) and chicken weight.

Cages cleaned, limed, and sprayed disinfectant. Equipment is cleaned and chaff sown in each plot enclosure. Randomization was performed prior to the placement of the cage broiler chickens by compiling numbers and repeat treatment prior to any cage that has been prepared, then selected at random. The feed is given appropriate treatment level from the first day until day 37. Feed and water given ad libitum.

To prevent the disease, the age of four days of ND vaccination via eye drops, and Gumboro vaccination by subcutaneous injection. Age of 28 days was stage II ND vaccine orally. Weight was measured every week in each treatment cage to determine weight gain in broilers treatment.

When chicken broiler aged 21 days, was the treatment in accordance with predetermined levels to determine the effect of papaya leaf flour. When 34-day-old broilers, weighing is done final body weight (g). Furthermore each experimental units drawn at random each animal and transferred to 24 cages each cage measuring 50x60 cm<sup>2</sup>. At the bottom of the cage prepared excreta plastic container measuring base enclosure.

In sampling, the results of excreta that exist in each treatment cage, put it in a plastic container, mixed evenly. Minimal excreta taken is weighing 100 grams and all plastic tied to the incoming air. After the sample is already in the next get level examination NH<sub>3</sub> and weight.

### Spectrophotometric method

Ammonia bound in boric acid 0.1% (can be replaced with sulfat acid) were analyzed with Nessler method (Spectroscopy Laboratory, 1992) as follows:

1. Determination of the absorption wavelength and the manufacture of ammonia gas calibration curve. Calibration curve made with different content of the mother liquor ammonia gas (0, 0.5, 1, 2, 4; 8; 16) plus 0.5 ml Nessler reagent. Then the absorbance was measured at a wavelength of 400 nm and searched the regression equation.
2. Ammonia gas analysis. Ammonia gas is bound in 0.1% boric acid mixed with 0.5 ml of Nessler solution that will produce a yellow to reddish-brown color. Color formed was measured with a spectrophotometer absorption at a wavelength of 400 nm.

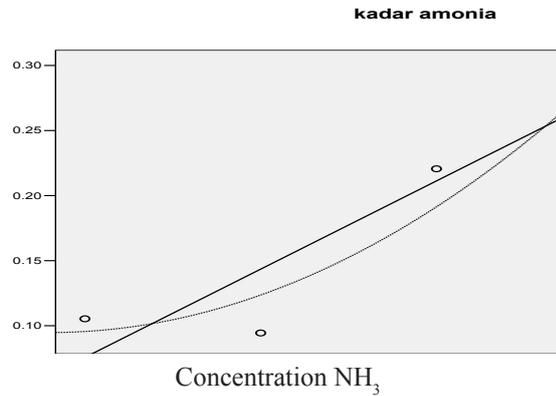
## Results

Obtained from the graph that the visible results of the difference decreased levels of ammonia (NH<sub>3</sub>) in the treatment of the addition of flour papaya 5% (P1) of the control (P0), but it happens raising levels of ammonia on the addition of flour treatment papaya 7% (P2), and the rising in addition of flour treatment papaya 9% (P3).

Table 1. NH<sub>3</sub> level examination results

The result of the addition of flour papaya levels	(%)
P0	0.1053
P1	0.0944
P2	0.2206
P3	0.2893

Description: P0 = group without papaya leaf; P1 = group receiving 5% papaya leaf; P2 = group receiving 7% papaya leaf; P3 = group receiving 9% papaya leaf



caption: ° observed, \_\_\_ linear, quadratic ----  
Figure 1. Graph NH3 levels.

Table 2. Average examination weight results (gram)

Treatment	Day	21 days	28 days	34 days
0%		853,3333	1355	1698,333
5%		850	1391,667	1811,667
7%		831,6667	1326,667	1695
9%		853,3333	1276,667	1596,667

Description: P0 = group without papaya leaf; P1 = group receiving 5% papaya leaf; P2 = group receiving 7% papaya leaf; P3 = group receiving 9% papaya leaf

In calculating the weight of chicken, in the calculation of statistics there are real differences in body weight. Feeding with the addition of papaya leaf flour 5% have tendency to be higher than others.

## Discussion

### Examination Levels of Ammonia (NH3).

Result showed that 5% papaya leaf in the ration tendency to decrease ammonia compared to the control. Meanwhile treatments with 7% and 9% of papaya leaf in the ration cause increasing of ammonia. This because of the treatment with 7% and 9% of papaya leaf flour reduced the palatability, so it caused decreasing of the papaya leaf intake.

So influential papaya leaf flour is also reduced. Thus the addition of flour giving papaya with levels of 5% (P1) is a tolerant and effective levels to lower levels of ammonia.

No statistically significant difference in decreased levels of ammonia (NH3) in the treatment of flour giving papaya 5% (P1) compared with controls (P0). This is due to the level of consumption and less repetition so it does not look real difference.

The result is supported from the calculation of the weight of chicken per week showed significant weight gain in the treatment of flour papaya adding 5% (P1) compared with controls (P0). According Suryana (2002), a high content of ammonia gas in chicken excreta, indicating less imperfect digestion or excess protein in the diet, so it is not absorbed nitrogen as amino acids, but released as ammonia in the excreta. Means the addition of flour papaya commission of 5% can improve the digestive process. Ammonia production is affected by a poor digestive system in chickens.

Rachmawati (2000) mentions that NH3, H2S, and CO2 gas often causes health problems for livestock, farmers, and the environment. Several studies on the effect of NH3 on poultry have been reported, which can reduce the average growth and feed efficiency, respiratory tract damage (Chronic Respiratory Disease) and increases the activity of ND virus (New Castle Disease). In other words, the addition of the provision flour papaya treatment that can improve the growth of body weight means reducing ammonia levels in broiler chickens excreta. NH3 concentration increases with the increase in moisture, pH, and temperature of the cage, as well as populations of microorganisms (Rohaeni, 2005). Papaya leaf flour can reduce production levels of NH3 in broiler excreta. According to Purnomo (2001) papaya flavonoids in the leaves have antibacterial activity against Staphylococcus sp, Propionobacterium and Corynebacterium sp. In the

flavonoid containing a phenolic compound. Phenol is an acidic alcohol that is also called carboic acid. Growth of *Escherichia coli* bacteria can be disrupted due to the presence of a phenolic compound contained in the ethanol extract of leaves papaya. Conditions by the presence of phenolic acids can affect to the reducing growth of bacteria *Escherichia coli*.

### **Weight examination**

In calculating the weight of chicken, in the calculation of statistics there are real differences in body weight. Feeding with the addition of papaya leaf flour 5 % increased weight gain compare to others. It shows the consumption of feed with the addition of 5% of papaya leaf flour is tolerant levels for broiler chickens. From the results showed highly significant results added.

## **Conclusion and Recommendation**

### **Conclusion**

1. Feeding chickens with the addition of flour papaya with levels of 5% tendency reduce levels of NH<sub>3</sub> on Broiler chicken excreta.
2. Feeding chickens with the addition of flour papaya with levels of 5% can increase the weight of broiler.

### **Advice**

In addition the use of flour papaya for Broiler chicken farm suggested 5% in chicken feed

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# Utilization of Haylage of Local Agro-Industrial Byproduct Pretreated with Afex Method

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## Abstract

The aim of this paper is to study the potential of haylage with pretreatment AFEX (ammonia fiber expansion) for ruminant feedstuffs. Agro-industrial by-products in Indonesia are very abundant and can be used as feedstuffs. High lignin content in these feedstuffs make them difficult to digest and cause inefficiency to support animal productivity. Accordingly, an effective and applicable technology is required to solve the respective problem. AFEX is a novel pretreatment process for cellulosic materials by combining concentrated ammonia, moderate temperatures and pressure for opening up biomass structure. This method can increase digestibility of fiber, increase in vitro digestibility by ruminal microorganism, increase crude protein digestibility and also increase of the productivity of animals. It may facilitate rumen microbes and the respective enzymes for the access into cellulose structure. Haylage is one of the forages preservation that can provide forages continuously every year. A combination AFEX treatment and fermentation like haylage is a good way to produce a ready to use feed and may be useful as feedstuff for ruminant.

**Keywords:** AFEX, agro-industrial by-product, fiber, haylage, ruminant

## Introduction

Indonesia has the potential feed sources that abundant on the agro-industrial sector. Mostly the availability of feed materials in Indonesia comes from byproduct and even waste product of agro-industrial. The majority of these materials contained fiber that was difficult to digest by animals. Lignin that has in byproduct of agro-industrial made microbes in rumen difficult to degraded it. Utilization byproduct and waste of agro-industrial as good as possible, it would be a good potential as a ruminant feed.

Several technologies have been used to enhance the fiber digestibility for agro-industry byproduct. Recently, ammonia fiber expansion (AFEX) pretreatment offers some benefits for ruminant animals. It is a novel pretreatment which can provide fibers such as cellulose depolymerization and partial solubilization of hemicellulose that easily to digest for ruminal microbes (Bals *et al.* 2010). Haylage is one of the good ways to continue AFEX pretreatment that can increase of palatability and also can supply forages continuously.

### The potential of Agro-industrial byproduct in Indonesia as fiber sources

Agro-industrial byproduct had a great potential in the sufficient availability of forages. Some of agro-industrial by product are rice straw, corn straw, corn stover, sugarcane bagasse, palm byproduct, and others. Syamsu *et al.* (2003) stated that the production of rice straw (85.81%) was the highest for agro-industrial byproduct followed by corn straw (5.84%) and peanut hay (2.84%). In West Sumatera, 1.75-2.1 million of animal units can be satisfied with the agro-industrial byproduct in that area (Table 1).

Table 1. The potential of the agro-industrial byproduct in West Sumatera at 2009\*

Commodities	Yield area (ha)	Yield (ton)	Agro-industrial byproduct (ton)	Potent for animals (livestock unit)
Rice	439,542	2,105,700	2.1 million tonnes of rice straw	572,343
Corn	70,882	404,795	283,528-345,410 tonnes of corn straw (4-5 t/ha)	155,358-194,197
Palm	170,092	363,904	1,071,579 ton palm fronds (6.3 t/ha/year)	595,322

\*Buharman (2011)

A low nutritive value of agro-industrial byproduct as ruminant feed due to the high fiber and lignin content. Fiber content in rice straw was 28.1% dry matter (DM), corn straw 24.4% DM, palm fronds 32.5% DM (Buharman 2011). The lignin contents in leave and frond of palm were 22-27% and 17-21%, respectively (Ginting 2012). The acid detergent fiber (ADF) and neutral detergent fiber (NDF) in each agro-industrial byproduct in corn straw were 29% and 48%, respectively and in palm byproduct were 48-54% and 75.6-83%, respectively (Ginting 2012; Umiyasih *et al.* 2008). Rice straw is the most widely used by farmers as forages for ruminant. Straws treated with urea and probiotics that feed to ruminant can increase of average daily gain, decrease of feed conversion rate and increase of feed efficiency compared with the control that feed elephant grass as a forages (Pritanti *et al.* 2001).

Ginting (2012) reported that sugarcane bagasse could be used as fiber sources for ruminant too. It has lower lignin content than palm fronds (10% vs 17-27%). In ruminants, the high lignin content can decrease digestibility and reduce animal productivity. Therefore, it is needed pretreatment process like hydrolytic, hydrothermal, and chemicals treatment for agro-industrial byproduct that have a possibility to improve the quality and quantity of animal product.

### **Pretreatment AFEX**

Several research about ammoniation in forages with high-fiber content have been carried out in Indonesia (Bata, 2008; Umiyasih & Wina, 2008; Ali, 2005). The advantages of ammoniation treatment are (1) effective preservative for hay containing up to 30% moisture, (2) increased the nutrient content of the feed, (3) increased the nitrogen content of feed, (4) improved palatability, intake and digestibility of feed (Bata, 2008; Umiyasih and Wina, 2008; Ali, 2005; Ahmed *et al.*, 2002). High pH in this process would breakdown of glycosides bond in the cellulose that making fiber was easier to digest for rumen microbes (Umiyasih and Wina, 2008).

One of the new technologies that have been developed by means of feed technology is ammonia fiber expansion (AFEX) method. This method is a novel pretreatment process for cellulosic materials by combining highly concentrated ammonia, moderate temperatures and pressure for opening up biomass structure. Application of AFEX method to highly fibrous feedstuffs results in a cellulose depolymerization and partial solubilization of hemicelluloses. It may facilitate rumen microbes and the respective enzymes for the access into cellulose structure. Furthermore, AFEX treatment may dramatically increase the rate and extent of both glucan and xylan release during enzymatic hydrolysis, which in turn may improve digestibility and crude protein content per unit of dry matter (Garlock *et al.* 2012; Bals, *et al.*, 2010). Thus in the future this pretreatment technique will be more effective and applicable to degrade fiber compared to conventional ammonia treatment. Subsequent feed technology such as fermentation will be the next treatment in order to produce a ready to use feed like haylage.

### **Post Treatment: Haylage**

Preservation of forage can be done by making silage, haylage and hay. Haylage has moisture content between hay and silage. Haylage produces lactic acid and water soluble carbohydrate. The presence of lactic acid in the haylage can reduce the pH into 4.8 and 5.11 (Kung Jr. *et al.*, 1983). This condition can inhibit the organisms growth that can damage the forage feed. Crude protein on haylage treatment that was resulted in swamp forage at South Kalimantan was greater than hay and silage treatments (Rostini *et al.*, 2014). Kume grass (*Sorghum Timorense*) haylage in East Nusa Tenggara added *Rhizopus oligosporus* enhanced crude protein and decreased crude fiber content in the feed (Hau and Nenobais, 2007).

Adding ammonia at high dry matter levels of alfalfa silage stimulated to increase the production of lactic acid, increase nitrogen in feed, improve insoluble nitrogen, decrease free amino acids compared to the control and microbe inocula addition. The addition of ammonia to haylage also can inhibit proteolysis by reducing the activity of degradative enzymes plant (Kung Jr. *et al.* 1983). Indirectly, it showed that AFEX pretreatment might be has a role in improving the quality of the forage through the process of haylage. Improvement of the efficiency of feed nutrient use in ruminant could be happened with the combination of AFEX pretreatment and haylage in forages.

### **The Potential of AFEX – Haylage Combination**

Agro-industrial byproduct in Indonesia was abundant which predominantly containing high fiber and lignin such as rice straw, corn straw, sugarcane bagasse and so on. It can be utilized as a source of forage for livestock, especially ruminants. AFEX pretreatment is suitable to be applied in the high fiber feed like agro-

industrial byproduct. Garlock *et al.* (2012) reported that corn stover with AFEX pretreatment reduced the NDF content and increased digestibility of NDF compared to untreated controls. The production of xylose, total sugar and also the amount of nitrogen in forages feed was increasing after gotten AFEX pretreatment (Bals *et al.* 2010). Rice straw was treated similar to AFEX enhanced milk production (Weimer *et al.* 2003).

The digestibility of NDF on corn stover AFEX treatment (593 g/kg of dry forage) was greater than ammonia treatment (512 g/kg of dry forage) and untreated (471 g/kg of dry forage). Increased NDF digestibility that were treated AFEX was occurred in rice straw (46%), corn silage (32%) and sugarcane bagasse (68%)(Bals *et al.* 2010). Application of haylage has been done at this tropical country. Goat that feeding haylage of swamp forage could increase crude protein digestibility, ether extract digestibility, and average daily gain (Rostini *et al.* 2014). Individually, AFEX treatment and haylage result a good effect in animal performances. So, there is a possibility of combination of AFEX and haylage was providing great results in sufficient of fiber source in ruminant and also enhancing quality and quality of livestock product.

It was caused on AFEX treatment the ester linkages within lignin and hemicelluloses were cleaved. Furthermore, during AFEX treatment oligomeric carbohydrates was formed and production of non-protein nitrogen increased. Then continuing to haylage as a preservation of forage could make multiple benefits in improvement feed quality. Because with AFEX and haylage combination can suppress proteolysis in forages thus ruminant can utilize N sources efficiently.

## Conclusion

Ammonia fiber expansion (AFEX) pretreatment is one alternative to breakdown lignin components that difficult to digested for rumen microbes. The AFEX pretreatment also increased crude protein content in feed. Preservation forages with haylage method can be applied in Indonesia. Combination of AFEX and haylage on agro-industrial byproduct with high crude fiber content was giving merits such as lifted up the availability of forages and also improved the livestock productivity.

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# Physical Quality and Storage Time Pellet *Indigofera* sp Leaves.

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## Abstract

This study conducted to find out the effect of different pellet diameter and storage time on physical properties of pelleted *Indigofera*-leaves. The research consist of two experiment; the first experiment was investigation of the die size (die 3, 5, and 8 mm) effects on physical properties usig a Completely Randomized Design (CRD) with 3 replications. The second experiment was the effect of storage quality on physical quality of pellets of the *Indigofera*-leaves. A factorial Completely Randomized Design (CRD) was applied with 3 replications, the first factor was die diameter consisting of 3, 5, and 8 mm and the second factor was the storage time (0, 7, 15, 30, and 60 days). The variables observed were physical pellet properties and pellet quality namely spesific gravity, bulk density, compacted bulk density, respone of angle and pellet durability index (PDI). The result of this experiment showed that specific gravity, bulk density, and compacted bulk density of the pellet are higher than *Indigofera* sp. leaves in mesh form (flour). The diameter difference of pellet do not significantly influence on the pellet durability index.

Keywords: *Indigofera* sp., Pellet diameter, physical quality, physical properties, storage

## Introduction

Development of feed industry should be supported by the production and supply of a continuous feed materials in quantity and quality. Production of feed material should be done efficiently at the level of economical production scale. Therefore, the study of the sources of potential feed ingredients to be produced on an industrial scale is very important in the context of supply chain management efficient feed. *Indigofera* sp. is a legume that is very good to be used as feed material as a source of protein. This legume has a high protein content and tolerant to drought, waterlogging and also resistant to salinity. In addition, this plant is easy to grow and has a biomass production of forage that is high enough.

Provision of plant *Indigofera* sp. as feed has disadvantages in terms of production (volume and storability) and distribution because it has a bulky and perishable nature so that the necessary processing technology so that the feed materials not only have good quality, but can be produced in large volumes and can be stored in less time long that is more efficient and available throughout the year.

Pellet-making process is a mechanical process using a combination of water vapor (moisture), heat, and pressure. Characteristics and particle size materials are two factors that affect physical endurance as well as the quality of pellets. Pellet process is processing technology that can be used to cultivate crops such as for age *Indigofera*. The aim of this study was to measure the physical properties and physical pellet quality of pelleted *Indigofera* leaves.

## Materials and Methods

The experiment was conducted in Feed Industry Laboratory, Department of Nutrition and Feed Technology. Fresh *Indigofera* leaves was harvested at 60 days of ages, dried up naturally under sun light up to the MC of 14%. The dried leaves was ground using Semi Fixed Hammer mill with 5 mm the screen size. As many as 15 Kg of ground leaves samples were processed using Pellet Presser. The research consist of two experiment; the first experiment was investigation of the die size (die 3, 5, and 8 mm) effects on physical properties usig a Completely Randomized Design (CRD) with 3 replications. The second experiment was the effect of storage quality on physical quality of pellets of the *Indigofera*-leaves. A factorial Completely Randomized Design (CRD) was applied with 3 replications, the first factor was die diameter consisting of 3, 5, and 8 mm and the second factor was the storage time (0, 7, 15, 30, and 60 days). The variables observed were physical pellet properties and pellet quality namely spesific gravity, bulk density, compacted bulk density, respone of angle and pellet durability index (PDI).

## Results and Discussions

### Physical characteristics of the pellets *Indigofera* sp. leaves

Physically pellet leaves *Indigofera* sp. has a color and odor are almost the same, namely dark green color and odor resembling fragrant tea leaves, but has a different texture. Pellet size 3 and 5 mm has a smooth texture and shiny, while the pellet size of 8 mm has a rough texture and seemingly less compact.

### The physical properties of the pellets *Indigofera* sp. leaves

The physical properties of leaves *Indigofera* sp. before and after the formed pellets have different characteristics, as shown in Table 1. Values density (BJ) leaves *Indigofera* sp. before the pellets formed is 601.61 kg / m<sup>3</sup>, whereas once formed pellets with size 3, 5, and 8 mm respectively increased to 1465.2 kg / m<sup>3</sup>, 1623.93 kg / m<sup>3</sup>, and 1674 kg / m<sup>3</sup>. The low value of BJ leaves *Indigofera* sp. before formed pellets show leaves *Indigofera* sp. the form of flour has properties bulky.

Table 1. Physical properties of *Indigofera* sp. leaves on powder and pellets

Variable	Powder	Pellet Size (mm)			Pellet Average
		3	5	8	
BJ (kg/m <sup>3</sup> )	606.61	1465.20	1623.93	1674.00	1587.86
KT (kg/m <sup>3</sup> )	290.33	620.71 <sup>A</sup>	625.41 <sup>A</sup>	567.97 <sup>B</sup>	604.69
KPT (kg/m <sup>3</sup> )	324.46	659.50 <sup>A</sup>	645.61 <sup>A</sup>	577.03 <sup>B</sup>	627.38
ST (°)	35.66	18.14 <sup>A</sup>	21.28 <sup>B</sup>	24.13 <sup>C</sup>	21.18
PDI (%)	-	97.91 <sup>A</sup>	96.09 <sup>B</sup>	90.86 <sup>C</sup>	94.95
Water Content (%)*	14.00	8.49	6.37	12.23	-

BJ = Density, KT = The Collision Density, KPT = Compaction Density Stack, ST = Stack Angle, PDI = Pellet Durability Index.

*Indigofera* sp. leaves powder has a density of pile (KTP) and compaction Density Stack (KPT) respectively 290.33 kg / m<sup>3</sup> dan 324.46 kg / m<sup>3</sup>. Once formed into pellets leaves *Indigofera* sp. increased density value of the stack and heap compaction density. KT value pellet size 3, 5, and 8 mm respectively was 620.71 kg / m<sup>3</sup>, 625.41 kg / m<sup>3</sup>, and 567.97 kg / m<sup>3</sup>, while the value of KPT row is 659.5 kg / m<sup>3</sup>, 645.61 kg / m<sup>3</sup>, and 577.03 kg / m<sup>3</sup>. Values pile density and compaction density pellets pile of *Indigofera* sp. leaves showed that the leaves of *Indigofera* sp. pellet form requires space or a smaller volume per unit of specific weight compared with leaves of *Indigofera* sp. flour form.

Value angle of repose (ST) leaves *Indigofera* sp. in powder (35,66°) higher than leaves *Indigofera* sp. pellet form. ST value pellet leaves *Indigofera* sp. 3, 5, and 8 mm in size are, respectively 18,14°, 21,28°, and 24,13°. This indicates that the pellet leaves *Indigofera* sp. has a much higher flow that would be more efficient in terms of handling, transport, and storage.

### The physical properties of the pellets *Indigofera* sp. leaves Diameter 3, 5 and 8 mm

Statistical analysis showed that the density was not affected ( $P > 0.05$ ) by the size of the die, which means the difference in the size of the die which is used does not change the value of BJ *Indigofera* sp. leaves. This is presumably because the size of the die 3, 5, and 8 mm is die size interval representing a uniform particle size in determining the specific gravity. Contrast Orthogonal test results showed that the pellet leaves *Indigofera* sp. size 3 and 5 mm KPT value KP and the same, but the value of both higher ( $P < 0.01$ ) compared with pellets *Indigofera* sp. size 8 mm.

Statistical analysis showed that there were significant differences ( $P < 0.01$ ) in die size difference of the value of ST. Further test results showed that the size of 3 mm pellets ST highly significant to the corner pile pellet sizes 5 and 8 mm, and corner pile pellet size of 5 mm highly significant to the corner piles of pellets the size of 8 mm. Angle stacks each pellet size is in the range below 30 ° which indicates that the pellets produced included into the group of materials that are very easy to flow on an inclined plane.

The statistical results showed that the die size difference was highly significant ( $P < 0.01$ ) to the value of the pellet durability. Further test results indicate that there are differences in PDI very real value between the size of 3 mm pellets with a pellet size of 5 and 8 mm, and a pellet size of 5 mm highly significant with pellets the size of 8 mm. All values above PDI pellets standard specification of minimum durability of 80%, ie to pellet the size of 3, 5, and 8 mm respectively 97.91%; 96.09%; and 90.86%, so the pellet leaves *Indigofera* has a value Pellet Durability Index (PDI) is good or not easily broken.

## The physical properties of the pellets *Indigofera* sp. leaves different sizes within the prescribed period

Results of analysis of variance showed that there were significant interaction ( $P < 0.05$ ) between the storage time and the size of the pellets to the value Pellet Durability Index (PDI). During the storage period, the average value of PDI decreased but there were no significant differences. The average value of PDI after the shelf life of 60 days showed values above the minimum value is in the range of 94.16 to 94.95%, which means that the pellets *Indigofera* sp. have a good shelf life even though stored for two months.

Results of analysis of variance showed that there were highly significant interaction ( $P < 0.01$ ) between the storage time and the size of the pellets on water content. The water content of the pellets during the shelf life tends to increase, despite a decrease in the shelf life of 30 days but increased back on the shelf life of 60 days. Results of variance showed that there is no interaction between the size of the pellet and the shelf life of the water activity, but the test results further indicate that the size of the pellets and the shelf life of highly significant ( $P < 0.01$ ) against water activity. The value of water activity on all treatments increased relative to the shelf life of 15 days and then slightly decreased hinga shelf life of 60 days. 8 mm pellet size has a water activity value higher than the pellet size 3 and 5 mm.

Based on the physical properties of green pellets *Indigofera* result pellet process produces significant physical changes, especially in terms the bulk density and the angle of repose values. Pelleted *Indigofera* sp. leaves has a lower volume of space thereby increasing efficiency in the store room and transport compared with plants that have not been made *Indigofera* sp pellet or powder form. The average value of PDI pellets *Indigofera* sp. is 94.95% indicates pellet forage *Indigofera* have good quality or not easily broken during handling. Leaves of *Indigofera* sp pellets also can be stored up to 60 days without showing significant changes in physical qualities (stable) thus possess a longer shelf life.

## Conclusion

Based on the physical properties of green pellets *Indigofera* result pellet process produces significant physical changes, especially in terms the bulk density and the angle of repose values. Pelleted *Indigofera* sp. leaves has a lower volume of space thereby increasing efficiency in the store room and transport compared with plants that have not been made *Indigofera* sp pellet or powder form. The average value of PDI pellets *Indigofera* sp. is 94.95% indicates pellet forage *Indigofera* have good quality or not easily broken during handling. Leaves of *Indigofera* sp pellets also can be stored up to 60 days without showing significant changes in physical qualities (stable) thus possess a longer shelf life.

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# Identification of Substrates of The Yeast Ubiquitin Ligase Rsp5 Under High-Temperature Stress Conditions

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## Abstract

*Rsp5 is an essential E3 ubiquitin ligase in Saccharomyces cerevisiae. This enzyme contains the membrane-bound C2 domain, three WW domains, which are the target protein interaction modules that bind proline-rich ligands, and the catalytic HECT-domain that ligates ubiquitin to the target protein. Previous studies have shown that, when yeast cells were exposed to stresses that induce protein denaturation, rsp5<sup>A401E</sup> mutant cells showed much more sensitivity to these stresses than the wild-type cells. This result suggests a novel function of Rsp5 as a key enzyme involved in selective degradation of stress-induced abnormal protein for yeast cell growth under stress conditions. Here we identified the substrate(s) of Rsp5 under high-temperature stress conditions by using yeast two hybrid system. Plasmids pGilda-Rsp5<sup>C777A</sup> HIS3, pJG4-5-cDNA TRP1 and pSH18-34 URA3 were used as a bait plasmid, a prey plasmid and a reporter plasmid, respectively. Yeast strains used in this study were BY4741 (a his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 Δtrp1 RSP5) and BY4741 rsp5<sup>A401E</sup> (a his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 Δtrp1 rsp5<sup>A401E</sup>). So far, we obtained three candidate genes, PGS1, ICT1 and BAG7, as substrates of Rsp5. PGS1 encodes phosphatidylglycerolphosphate synthase. ICT1 encodes lysophosphatidic acid acyltransferase, which is responsible for enhancing phospholipid synthesis, and is highly expressed during organic solvent stress. BAG7 encodes Rho GTPase activating protein (RhoGAP). RhoGAP stimulates the intrinsic GTPase activity of Rho1, which plays a role in actin cytoskeleton organization and control of cell wall synthesis.*

*Keywords: high-temperature stress, Rsp5, ubiquitin ligase, yeast two-hybrid*

## Introduction

The budding yeast *Saccharomyces cerevisiae* has been widely used in the fermentation industries to produce various useful metabolites, representatively ethanol and carbon dioxide. Under fermentation processes or industrial yeast production, yeast cells are exposed to various environmental stresses, such as high concentrations of ethanol, freezing, desiccation, and high osmotic pressure. These stress conditions induce several dysfunctions of proteins, including denaturation and misfolding (Gaseret *et al.*, 2008), generate reactive oxygen species (ROS) (Moraitis and Curran, 2004), and lead to growth inhibition or cell death. The degradation of a protein through ubiquitin system involves three types of different enzymes: (1) E1 enzymes known as ubiquitin-activating enzymes that modify ubiquitin into a reactive state, (2) E2 enzymes known as ubiquitin-conjugating enzymes that carry ubiquitin to E3 and the substrate proteins, and (3) E3 enzymes as ubiquitin ligases that play an important role in recognizing the substrate proteins (Hershko and Ciechanover, 1998). Among E3 enzymes, Rsp5 is the only yeast member of the highly conserved mammalian Nedd4 family of HECT-type E3 ligases (Tardiff *et al.*, 2013). Numerous studies demonstrated that Rsp5 is an essential ubiquitin ligase in *S. cerevisiae* involved in various cellular functions, including endocytosis, multivesicular body sorting, RNA export, transcription, lipid biosynthesis, mitochondrial inheritance, and protein catabolism (Belgareh-Touze *et al.*, 2008; Shearwin-Whyatt *et al.*, 2006). The ability of Rsp5 to act as a multifunctional E3 in yeast is, at least in part, due to its capacity to modify different substrates with distinct mono- and poly-ubiquitin signals. Several proteins that function in endocytosis, such as Rvs167, are monoubiquitinated in an Rsp5-dependent manner (Stamenova *et al.*, 2004). In contrast, Rsp5 targets a number of cellular proteins for polyubiquitination, including the large subunit of RNA polymerase II, the vacuolar membrane protein Sna3, and the mRNA nuclear export factor Hpr1 (Chang *et al.*, 2000; MacDonald *et al.*, 2012). The yeast two hybrid system is one of genetic method to identify protein-protein interaction. However, Rsp5-substrate interactions are still difficult to identify by the two-hybrid system. Such interactions may not be maintained to levels sufficient for their detection because of the ubiquitin-dependent degradation of substrates. We therefore reasoned that Rsp5 - substrate interactions could be identified by designing a two-hybrid system to inhibit the degradation of substrates during screening. In

this study we tried to get more understanding of the role of Rsp5 to recognize normal protein and degraded protein under temperature-stress condition, and to identify new substrate of Rsp5. Furthermore we will study the mechanisms how Rsp5 regulate ubiquitination for degradation of its substrates under stress condition.

## Materials and methods

### Screening for Potential Substrates for Rsp5-Dependent Degradation

The DupLEX-A Yeast Two-Hybrid System (OriGene) was used to establish the two-hybrid system for isolation of Rsp5-substrate interactions. Bait and prey were expressed under the control of *GALI* promoter in pGilda (OriGene) and pJG4 -5 (OriGene), respectively. pSH18 -34 (OriGene) was used as the reporter gene (LacZ) plasmid. After obtaining the transformants, they were transferred by placing nitrocellulose filters on the surface of the plates containing the transformants, and then the filters were pulled off and placed yeast-side up on plates that contained glucose and galactose (as carbon source) to induce the expression of bait and prey. The plates also contained X-gal for the detection of LacZ expression. Cells on the filters were incubated at 25°C for 12 h, and then at 37°C for 3–10 h until color development was evident. A total of 65 colonies were identified, and pJG-based plasmids were isolated from each colony. These plasmids were retransformed into yeast BY4741  $\Delta trp\ rsp5^{A401E}$  mutants harboring pGilda-rsp5<sup>C777A</sup> and pSH18 -34. As control, pJG-*BUL1* was also transformed into BY4741  $\Delta trp\ rsp5^{A401E}$  mutants harboring pGilda-rsp5<sup>C777A</sup> and pSH18-34. Colonies on each plate were tested for their abilities to activate LacZ gene. We selected 18 clones whose transformants turned blue uniformly, when coloration was evident.

### Western blot analysis

Yeast cells were cultured to the mid-log growth phase in SC-His/Ura/Trp liquid medium. Approximately  $10^7$  cells were harvested by centrifugation. Pelleted cells were disrupted with glass beads in 10% trichloroacetic acid. Supernatants obtained after centrifugation for 5 min at  $3000 \times g$  were boiled in two-fold concentration of sample buffer (50mM Tris-HCl [pH 8.0], 2% SDS, 0.0125% BPB, and 2.25% glycerol) for 3 min at 95°C. One microgram of protein was then loaded onto a 15% SDS-polyacrylamide gel. The proteins were transferred to PVDF membranes and then blocked with 5% skim milk (wt/vol) in  $1 \times$  TBST buffer (20 mM Tris-base, 150 mM NaCl, hydrochloric acid to neutralize the solution to pH 7.6 and 0.1% Tween-20). Proteins were reacted with an anti-HA mouse antibody (Roche) at 1:2,000 dilutions and RSP5 was detected by an anti-rsp5 mouse antibody (Invitrogen) at 1:10,000 dilutions as a primary antibody. An HRP-conjugated anti-mouse IgG was used as a secondary antibody at 1: 2,000 dilutions. The protein targets were detected by Pierce ECL Western Blotting Substrate (Thermo Scientific) and visualized using Fuji LAS4000 imager (GE Healthcare).

## Results and discussion

Although *BUL1* is a substrate of Rsp5, their interaction could not be detected by the conventional two-hybrid system, because it could be targeted to degradation by ubiquitin system. We therefore inhibit the degradation pathway during screening process. We used the bait plasmid harboring Rsp5 catalytically inactive mutant *rsp5*<sup>C777A</sup> and also we used Rsp5 mutants, which are defective in ubiquitination of substrates at their restriction temperature (37°C), as host cells for screening. After formation of transformant colonies, the two-hybrid interaction was monitored at 37°C.

Table 1. Detection of Rsp5-substrate(s) interaction by yeast two hybrid system

Bait	Prey	Host cell	Temp up-shift	LacZ activity
pGilda-Rsp5 <sup>C777A</sup>	pJG 4-5-BUL1	BY4741 <i>rsp5</i> <sup>A401E</sup>	+	+
pGilda-Rsp5 <sup>C777A</sup>	pJG 4-5	BY4741 <i>rsp5</i> <sup>A401E</sup>	+	-
pGilda-Rsp5 <sup>C777A</sup>	pJG 4-5	BY4741 <i>rsp5</i> <sup>A401E</sup>	-	-
pGilda-Rsp5 <sup>C777A</sup>	pJG 4-5-PGS1	BY4741 <i>rsp5</i> <sup>A401E</sup>	+	+
pGilda-Rsp5 <sup>C777A</sup>	pJG 4-5-PGS1	BY4741 <i>rsp5</i> <sup>A401E</sup>	-	-
pGilda-Rsp5 <sup>C777A</sup>	pJG 4-5-ICT1	BY4741 <i>rsp5</i> <sup>A401E</sup>	+	+
pGilda-Rsp5 <sup>C777A</sup>	pJG 4-5-ICT1	BY4741 <i>rsp5</i> <sup>A401E</sup>	-	-
pGilda-Rsp5 <sup>C777A</sup>	pJG 4-5-BAG7	BY4741 <i>rsp5</i> <sup>A401E</sup>	+	+
pGilda-Rsp5 <sup>C777A</sup>	pJG 4-5-BAG7	BY4741 <i>rsp5</i> <sup>A401E</sup>	-	-

The plasmids used: Bait : pGilda-Rsp5<sup>C777A</sup> HIS3; Prey : pJG4-5-cDNA TRP1 Reporter plasmid : pSH18-34 URA3. The yeast strain used : BY4741 (*a his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 Δtrp1 RSP5*), BY4741 *rsp5*<sup>A401E</sup> (*a his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 Δtrp1 rsp5*<sup>A401E</sup>) and CKY8 (*a ura3-52 leu2-3,112*)

From ~30.000 independent colonies, we obtained 18 transformants with blue coloration uniformly. We retransformed to *E. coli* DH5α cells and re-introduced to *S. cerevisiae* *rsp5*<sup>A401E</sup> cells to test the plasmid dependency. Three strong candidate genes of Rsp5 substrate under high-temperature stress were *PGS1*, *ICT1* and *BAG7* as shown in Tabel 1. *PGS1* encodes Phosphatidylglycerolphosphate synthase, catalyzes the synthesis of phosphatidylglycerolphosphate from CDP-diacylglycerol and sn-glycerol 3-phosphate in the first committed and rate-limiting step of cardiolipin biosynthesis. *ICT1* encodes Lysophosphatidic acid acyltransferase, responsible for enhanced phospholipid synthesis during organic solvent stress; null displays increased sensitivity to Calcofluor white; highly expressed during organic solvent stress. *BAG7* encodes Rho GTPase activating protein (RhoGAP), stimulates the intrinsic GTPase activity of Rho1p, which plays a role in actin cytoskeleton organization and control of cell wall synthesis; structurally and functionally related to Sac7p.

Next we checked physical interaction of Rsp5 – substrates using co-immunoprecipitation. To get more protein, we overexpress the candidate of Rsp5 substrates by GAL1 promoter under normal and high temperature stress. We used genome integration system to put HA-tagged protein in the genome of yeast. GAL1 promoter is known as a strong promoter was successfully over express all three candidate genes *PGS1*, *ICT1* and *BAG7* as shown in Figure 1A. The physical interaction of Rsp5 and its substrates is shown in Figure 1B.

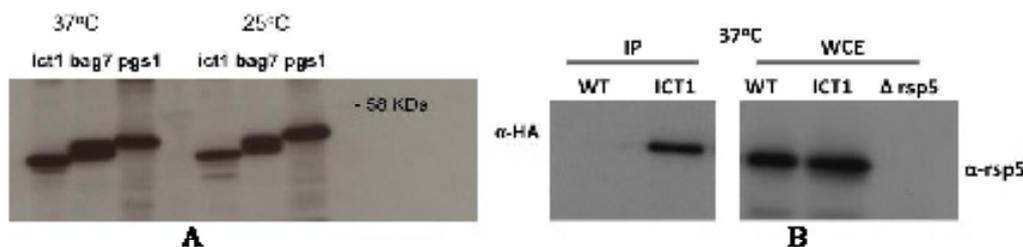


Figure 1. A. Overexpression of Rsp5 candidate substrates; B. Co-immunoprecipitation of Rsp5-ICT1 under high-temperature stress conditions (37°C). Yeast strain: BY4741 *Mata his3Δ1; leu2Δ0; met15Δ0; ura3Δ0; Δtrp1* and BY4741 *rsp5*<sup>A401E</sup> *Mata his3Δ1; leu2Δ0; met15Δ0; ura3Δ0; Δtrp1*

The physical interaction only shown in *ICT1* protein as a strong candidates of Rsp5 substrate. We need to understand what is the physiological function of this interaction in yeast cell under high-temperature stress.

## Conclusion

*ICT1* is the strong candidate of Rsp5 substrate under high-temperature stress condition. The physiological role of its interaction is still unclear.

## Acknowledgment

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# Feeding Wafer For Sheep

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## Abstract

*Thin-tailed sheep is one of local sheep that can support the needs of people's animal protein. Wafer complete feed of waste vegetable market is one of the feed results of technology that have nutritional value better than feeding a conventional feed i.e. forage and rice bran. This research was conducted at Laboratory of Feed Industry, Faculty of Animal Science, Bogor Agricultural University, Indonesia. The acceptability, performance test were conducted at Gapoktan Farm, Cilangkap-Jakarta, on July-November 2013. Experimental design used randomized block design with 5 treatments and 3 replications. The treatments were wafer feed composition i.e R1 (100% of conventional feed), R2 (75% of conventional feed+ 25% of wafer feed), R3 (50% of conventional feed + 50% of wafer feed), R4 (25% of conventional feed + 75% of wafer feed), R5 (100% of wafer feed). The results in this study indicated that the addition of water on wafer had significantly different ( $P<0.05$ ) on acceptability of sheep at 3 and 6 weeks storage. Wafer of feed were increase the sheep's final body weight, but it didn't have any effect on sheep's daily consumption. Wafer of feed that was given 100% to the sheep had the lowest feed conversion. Meanwhile, level of 25% of wafer complete feed had the highest value of IOFC.*

*Keyword: acceptability, body weight, feed conversion, sheep, wafer*

## Introduction

Sheep population in East Jakarta are 1744 head, this will result the increasing of feed requirements (Central Bureau of Statistics, 2012). Scarcity of forage has caused farmers to utilized waste vegetable from the market as their livestock feed. One of the sheep in East Jakarta is called the thin tail sheep. This sheep has characteristics of short tail and small body, its hair color is generally white, coarse and irregularly in the body (Arifin et al., 2007). As it is known that forage productivity is seasonal. During the rainy season, forage stock is abundant, but during the dry season forage stock is only a few or even none so that the sheep productivity will be decreased. Sheep farms are relied heavily on forage productivity that determine succeed of the farm. In order to solve these problems, it needs to look for alternative feed forage in the dry season. Vegetable waste when it is used as a raw material has several advantages that have economic value because it can produce a variety of useful products and easily obtainable, cheap, and available, also can reduce the problem of environmental pollution caused by waste (Retnani et al., 2014). The weakness of this vegetable waste is easy to decay, voluminous (bulky) and the availability is fluctuated, so the processing technology is needed to make this vegetable waste to be durable, easy to stored and easy to given to the animal. In order to solve this problem is by making vegetable waste into wafer feed. A pressing technology can make feed product into a wafer form. The wafer feed must contain energy; mineral; vitamin and protein needed by animal to increase productivity (Retnani et al., 2010a).

## Materials and Methods

The experiment used 15 thin heads sheep with average initial body weight around 27.43±5.43 kg. The experimental sheep were maintained individually. The ratio used consisted of two types conventional feed (field grass and rice bran) and wafer feed. Nutrient composition of wafer feed (% Dry Matter) is presented on Table 1.

Table 1. Nutrient Composition of Wafer Feed (% Dry Matter)

Wafer feed	Water Content	Ash	Crude protein	Crude Fiber	Crude fat	NFE
Nutrien	15.79	12.41	16.90	23.04	4.18	43.47

Laboratory Analysis of Feed Science and Technology (2012)

Figure 1 showed that diagram process of wafer feed production by chopping, drying, mixing, pressing, heating and forming with temperature 100°C for 10 minutes to get wafer feed and then cooling in room temperature (Retnani *et al.*, 2014).

### Experimental Design

The experimental design used randomized block design with 5 treatments and 3 replications. The treatments were wafer feed composition i.e R1 (100% of conventional feed), R2 (75% of conventional feed+ 25% of wafer feed), R3 (50% of conventional feed + 50% of wafer feed), R4 (25% of conventional feed + 75% of wafer feed), R5 (100% of wafer feed). Conventional feed were field grass and rice bran. The data were analyzed with the analysis of variance, and the differences among treatments were examined with orthogonal contrast test (Steel and Torrie, 1993). Wafer feed variables measured were acceptability, body weight, feed consumption and feed conversion.

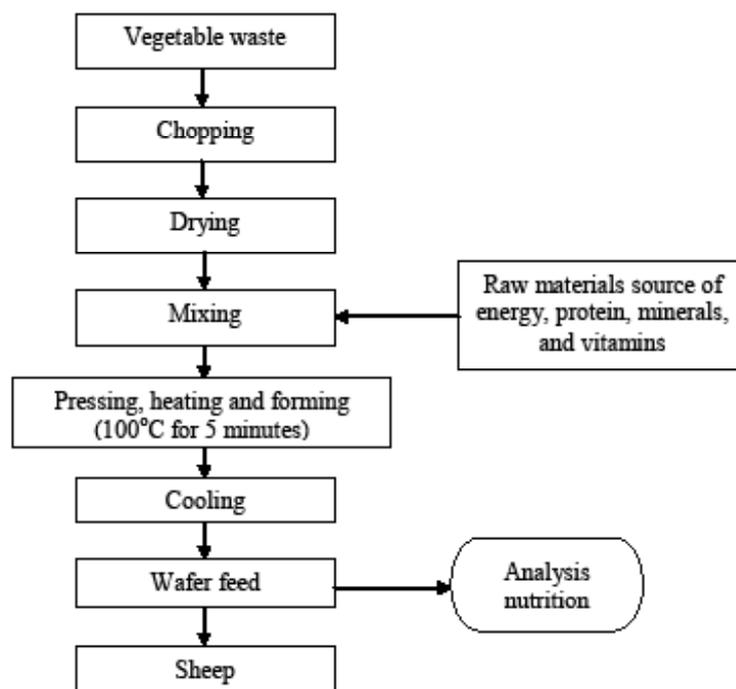


Figure 1. Diagram Process of Wafer Feed Production (Retnani *et al.*, 2014)

### Results and Discussion

Acceptability can be interpreted as an acceptance of livestock against a given feed (Stewart *et al.* 1998). The addition of water at 0 weeks of storage did not affect the acceptability of sheep on the wafer complete feed. Wafers which were not added with water had the lowest value of acceptability about  $18.30 \pm 23.86$  g or 0.025% BB, and the value of the highest acceptability by the addition of water was as much as 75% of the weight of the feed given around  $66.98 \pm 38.08$  g or 0.093% BB.

Final body weight in this study ranged from 27.07-34.00 kg/head. The result showed that wafer of feed treatment could increase final body weight of sheep. Treatment of R1 (100% of conventional feed) have final body weight was  $27.07 \pm 6.87$ , R2 (75% of conventional feed+ 25% of wafer feed) is  $32.87 \pm 4.91$ , R3 (50% of conventional feed + 50% of wafer feed) is  $32.07 \pm 10.16$ , R4 (25% of conventional feed + 75% of wafer feed) was  $29.53 \pm 6.12$  R5 (100% of wafer feed) was  $34.00 \pm 1.00$ . The treatment of R5 (100% wafer feed) has average body weight of the highest compared to other treatments. Sheep were fed by conventional feed had final body weight 27.07 kg, meanwhile sheep were fed 100% of wafer feed complete had 34 kg or 25.6% higher than conventional. According to Purbowarti *et al.* (2005). body weight thin tail sheep can reach 30-40 kg in males.

The result showed that wafer of feed treatment did not significant ( $P > 0.05$ ) on sheep's daily consumption. Treatment of R1 (100% of conventional feed) had sheep's daily consumption was  $1559 \pm 97$ , R2 (75% of conventional feed+ 25% of wafer feed) is  $1598 \pm 156$ , R3 (50% of conventional feed + 50% of wafer feed)

was  $1624 \pm 117$ , R4 (25% of conventional feed + 75% of wafer feed) was  $1454 \pm 138$ , R5 (100% of wafer feed) was  $1487 \pm 109$ .

Feed conversion was affected by feed quality, digestibility value, and efficiency. Increase in feed quality will improve body weight gain, so feed conversion value will decrease, meaning that the application of feed is efficient (Pond *et al.*, 1995). Feed conversion depends on dry matter intake and body weight gain. Feed conversion in this study ranged from 9.19-38.50.

Income is one of the main objectives in farm. By knowing the amount of income received by then a farmer can determine if feed costs incurred during the maintenance of livestock or not economical enough. IOFC (Income Over Feed Cost) that calculates the difference between sheep sales revenue minus feed cost incurred during the maintenance process.

The amount of benefits obtained by calculating the value of the business efficiency is the difference between sheep sales revenue minus feed cost incurred during the process of maintenance. IOFC were Rp. 10.100 (R1), Rp. 164.100 (R2), Rp. 156.800 (R3), Rp. 31.400 (R4), Rp. 146.050 (R5). The highest Income Over Feed Cost feed of sheep fed with 25% of wafer feed was Rp. 164.100,-.

## Conclusion

Wafer of feed were able to increase the sheep's body weight, but didn't effect on sheep's daily consumption. Wafer of feed that was given 100% to the sheep had the lowest feed conversion. Meanwhile, level of 25% wafer complete feed had the highest value of IOFC.

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**THEME C.**  
**FORAGE PRODUCTION AND TECHNOLOGY**

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# Potential of Dwarf Elephant Grass (*Pennisetum purpureum* Schum. cv. Mott) in Dry Land Areas of Bojonegoro as Forage-Based Feed Sustainability

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## Abstract

Animal industry especially stockers needs forage as their fibre and protein resource to maintain its quality and productivity. However, in several dry climate area, forage production is still low when it is dry season. So, the study about forage is very urgently and importantly needed. This study was conducted to evaluate the potency of dwarf elephant grass (*Pennisetum purpureum* Schum. cv. Mott) that has been planted on dry land areas of Bojonegoro. This research consists of two phase. The first phase of this research carried out on green house to evaluate the impact of drought stress by using 16 dwarf elephant grass, four water level of treatment (P1=100% KL; P2=75% KL; P3=50% KL; P4=25% KL) in completely randomized experimental design. The result showed that *Pennisetum purpureum* Schum. cv. Mott survived in those treatment without decreasing their dry matter (DM) productivity ( $P < 0.01$ ). It means that *Pennisetum purpureum* Schum. cv. Mott can be planted on drought areas of Bojonegoro. The second phase of this research carried out on dry land areas of Bojonegoro by using 320 dwarf elephant grass. Grasses were panted by using 2 x 5 completely randomised design. The first factor was the location (opened area and teak forest) and the second factor was the five fertilizer level (LEISA model). The result showed that both of two factor affected the DM production ( $P < 0.01$ ). The correlation between two factors also affected the DM production ( $P < 0.01$ ). The best result of planting the *Pennisetum purpureum* Schum. cv. Mott using 100 % anorganic fertilizer (Urea, SP36, and KCl) ( $P < 0.01$ ). This research conclude that *Pennisetum purpureum* Schum. cv. Mott has capability to be planted on dry land areas as forage-based feed sustainability.

**Keywords:** dry matter, drought stress, forage-based feed sustainability, LEISA, *Pennisetum purpureum* Schum. cv. Mott

## Introduction

Feed is one of main factor for livestock. One of ruminant feed is forage in a good quality contained protein, energy, vitamin and mineral (Herlina, 2003). However, Indonesia as a tropical country, has varied climates that affect forage production. Quantity, quality, and continuity of forage affects the ruminant productivity (Widiati, 2003). Dwarf elephant grass (DEG) (*Pennisetum purpureum* Schum. cv. Mott) has high productivity level. On rainy season, it has 2-2.5 tons/ha and 1.6 tons/ha on dry season (Olivo *et al.*, 1992 and Coser *et al.*, 1997). It means that DEG has a potency as drought resistant plant.

Bojonegoro is one of regency on east java that has high amount of ruminant population. On 2013, it has 160 037 cows, 19 dairy, 1 026 buffalo, 105 013 goats, 129 990 sheeps (BPS Bojonegoro, 2013). However, Bojonegoro has a long drought season. It has monthly average rain interval of 142 mm that classified as drought areas (Goenadi, 2003). Therefore Bojonegoro needs drought resistant forage to fulfill the ruminant needs. This research aimed to get several informations about the potency of DEG as drought resistant forage. Furthermore, it has purpose to get information about DEG as a part of integrated planting system between forage and teak forest.

## Materials and Methods

This research consists of two stages. The first stage take place on green house of agrostology unit, IPB, starting on October to December 2014. And the second stage take place on Sambeng, Bojonegoro, starting on December 2014 to January 2015.

## First stage

This study was carried out to evaluate the drought respon of DEG with four water level treatments (25%, 50%, 75%, and 100% field capacity). The treatments used completely randomized design and four replication. Each of them planted on 5 kg polybag that contained 10% of manure. The field capacity determined by using freely drainage model (Jury *et al.*, 2001). The first stage observed the morphological respons (height of plant, diameter of plant, root weighth and amount of leaves), physiological respons (water consumption, level of relative water content, and dissolved sugar level), and nutrientcontent (crude fibre and crude protein).

## Second stage

This study was carried out to evaluate the productivity of DEGthat planted in Bojonegoro. The treatments used competely randomized design. The location consist of opened area and integrated area (teak). There were five fertilizer level and composisiton. They were 100% organic, 75% organic and 25% anorganic, 50% organic and 50% anorganic, 25% organic and 75% anorganic, 100% anorganic. Anorganic fertilizer consist of Urea, SP36, and KCl. The fertilizing method using LEISA model (Low Eksternal Input Sustainable Agriculture) to get the lowest fertilizer input but earn the highest productivity (Giovannucci, 2007). After harvesting, the biomass were analyzed to get nutrient information by Proximate (AOAC, 2005) and Van Soest (1991).

## Result and Discussion

### First stage (drought respons of *Pennisetum purpureum* Schum. Cv. Mott)

#### *Morphological responses*

Morphological response of *Pennisetum purpureum* Schum. Cv. Mott observed height, diameter, root weight, and number of leaves. Drought stress treatment significantly affected ( $P < 0.05$ ) on plant height and number of leaves of the plant, excepted the plant diameter. Plant height of P1 treatment showed the best response. Water used by the plant for cell metabolism process (Liu and Stutzel, 2008). The number of leaves P1, P2, and P3 showed the best response. However, the quantity of leaves P1 was less than P3 due to the adaptation to defend themselves from the drought. Liu and Stutzel (2002) stated that in orderto response drought stress, the lower the rate of growth to reduce the rate of water loss. Weight roots influenced ( $P < 0.05$ ) by treatment of water stress. Root weight at P1, P2, and P3 showed a greater response than P4. However, at average, P1 showed greater results than P2, and P2 was greater than P3. This was caused by the growth of roots in drought stress tends to be greater to find the source of water. On the condition of drought stress, plants tend to decrease the production of biomass and increase the production of roots to find water sources (Yin *et al.*, 2005). In P4 treatment showed that no growth of roots due to the water concentration in soil was sufficient so water coming into the plant tissues by diffusion.

Table 1. Morphological respons of *Pennisetum purpureum* Schum. cv. Mott

Parameter	Treatment			
	P1	P2	P3	P4
Plant height (cm)	106.00±5.00c	83.37±17.81b	107.00±7.21bc	4.70±0.81a
Stem diameter (cm)	2.00±0.26	1.86±0.32	1.89±0.24	1.79±0.09
Amount of leaves	4.00±1.71b	4.00±2.40b	5.00±1.73b	1.00±0.50a
Root weight (gram)	0.38±0.13b	0.35±0.09b	0.30±0.19b	0.00±0.00a

Values with different letters differ significantly within column (upper case) or within line (lower case) at  $P < 0.05$ . P1:25% field capacity; P2:50% field capacity; P3:75% field capacity; P4:100% field capacity.

#### *Physiological responses*

Drought stress affects plant physiology mechanism to maintaining itself. Table 2 shows that the drought stress on *Pennisetum purpureum* Schum. cv. Mott significantly different ( $P < 0.05$ ). The increasing of water capacityin soil media would increasedthe leaf relative water content. Drought stress treatment has no significant effect ( $P > 0.05$ ) on water consumption. Drought stress affected on total production of sugar dissolved on stem. Nofyangtri (2011) stated that the physiological response of stressed plants showed a

decline in value of relative water content of leaves. Otherwise, total dissolved sugar would increased when drought stress level increased.

Table 2. Physiological respons of *Pennisetum purpureum* Schum. cv. Mott

Parameter	Treatment			
	P1	P2	P3	P4
Relative water content	13.30±2.66a	79.88±27.24b	84.89±6.46b	91.73±6.76b
Dissolved sugar level	2.25±1.5b	1.75±1.92ab	1.65±0.3ab	0.00±0.00a

Values with different letters differ significantly within column (upper case) or within line (lower case) at P<0.05.

P1:25% field capacity; P2:50% field capacity; P3:75% field capacity; P4:100% field capacity.

### Nutrient contents

Range of crude protein of *Pennisetum purpureum* Schum. cv. Mott is 15.40% to 21.94%. At the same time crude fiber content ranging from 22.51% to 25.77%. Crude protein content in P1 to P4 decreased respectively but the crude fibre in P1 to P4 increased respectively. Lutfi (2000) stated that nitrogen intake from the soil would increased the protein content of plant and phohibit the celulose and hemi-celulose on plant cell wall.

Table 3. Nutrient content of *Pennisetum purpureum* Schum. cv. Mott

Parameter	Treatment			
	P1	P2	P3	P4
Crude protein %	21.94	19.94	16.06	15.40
Crude fibre %	23.24	24.99	22.51	25.77

Analyzed by Pusat Antar Universitas (PAU) laboratory (IPB 2015)

P1:25% field capacity; P2:50% field capacity; P3:75% field capacity; P4:100% field capacity.

### Second stage (forage integrating system of *Pennisetum purpureum* Schum. cv. Mott)

The planting of *Pennisetum purpureum* Schum. cv. Mott along 40 days on Bojonegoro showed different result between opened field and integrated field.

### Morphology responses

*Pennisetum purpureum* Schum. cv. Mottthat planted by integrated model with teak forests (Table 4) showed the greatest response rates to treatment with 100% inorganic fertilizers (N5) in the production of dry weight. Whitehead (2000) stated that nitrogen was essensial element for plant growth.

Table 4. Morphology of *Pennisetum purpureum* Schum. cv. Mott planted by integrated system with teak

Parameter	N1	N2	N3	N4	N5
Plant height (cm)	95.50	98.88	99.38	92.00	86.88
Dry matter production (cm)	31.43a	30.13a	44.40ab	83.88b	117.00c

Values with different letters differ significantly within column (upper case) or within line (lower case) at P<0.05.

N1:100% organic fertilizer; N2:75% organic and 25% inorganic fertilizer; N3:50% organic and 50% inorganic fertilizer; N4:25% organic and 75% inorganic fertilizer; N5:100% inorganic fertilizer.

*Pennisetum purpureum* Schum. cv. Mottplanted in open fields (Table 5) showed the greatest response rates to treatment with a combination of 75% fertilizer organic + 25% inorganic (T4). However, for the production represented by the largest of dry weight in the treatment of 100% inorganic fertilizers (T5). Whitehead (2000) fertilizer that content nitrogen would increased the plant produktivity.

Table 5. Morphology of *Pennisetum purpureum* Schum. cv. Mott planted on opened field

Parameter	T1	T2	T3	T4	T5
Plant height(cm)	62.00	61.13	62.13	76.00	73.38
Dry matter production (cm)	172.93a	234.30b	379.63c	185.15a	369.98c

Values with different letters differ significantly within column (upper case) or within line (lower case) at P<0.05.

T1:100% organic fertilizer; T2:75% organic and 25% anorganic fertilizer; T3:50% organic and 50% anorganic fertilizer; T4:25% organic and 75% anorganic fertilizer; T5:100% anorganic fertilizer.

## Nutrient contents

Nutrient analyzed only occurred on highest response of morphology of *Pennisetum purpureum* Schum. cv. Mott. The highest result of morphology showed on 100% inorganic fertilizer treatment on integrated field and open field. The analyzed to fulfill the nutrient information of the highest productivity level of DEG.

Dry matter and crude fibre at N5 treatment in the integrated field has lower value than open fields. However, the value crude protein of N5 in the integrated field has higher value than open fields. Quality of fibre fraction (Van Soest, 1991) in N5 treatment on integrated field has lower value of ADF, NDF and cellulose than open field. However, lignin value on integrated field was higher than on open field.

Table 6. Nutrient content of *Pennisetum purpureum* Schum. cv. Mott

Parameter	Integrated field (N5)	Open field (T5)
Proximate (%)		
Water content	90.53±0.16	93.22±0.16
Ash	16.53±0.69	24.80±2.07
Crude protein	5.20±0.03	3.86±0.13
Crude fat	1.91±0.45	0.64±0.13
Crude fibre	27.09±0.87	29.69±1.33
Van Soest (%)		
ADF	36.64±2.01	47.67±2.06
NDF	58.15±0.36	61.87±0.25
Celulose	16.96±2.56	30.91±0.70
Lignin	19.78±2.50	16.76±1.40

Analyzed by Feed Biotechnology Laboratory of LIPI (2015)

## Conclusion

*Pennisetum purpureum* Schum. cv. Mott has potency to be planted on drought area to fulfill the ruminant forage need. *Pennisetum purpureum* Schum. cv. Mott also has a good morphologic and physiological responses to drought stress. By using LEISA model, *Pennisetum purpureum* Schum. cv. Mott gave the best growth respond on integrated fields.

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# Development of *Indigofera zoolingeriana* and *Pueraria javanica* on Dry Land Integrated with Teak Forest in Bojonegoro

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## Abstract

Forage needed by ruminant to reach optimum production of meat especially in tropical areas such as Indonesia. Therefore good quality and quantity production should be developed to supply that requirement. This study was aimed to develop *Indigofera zoolingeriana* and *Pueraria javanica* that integrated with teak forest in dry land of Bojonegoro. This study was conducted during the period of September 2014 to April 2015. This study consist of two phase. The first phase conducted on green house to evaluate the adaptability of *Indigofera z.* and *Pueraria javanica* in drought stress. The result showed *Indigofera z.* and *Pueraria javanica* both adaptive in 100% field capacity (cp), 75% cp, 50% cp, 25% cp, without decreasing their dry matter (DM) productivity ( $P < 0.01$ ). The second phase conducted on dry land bojonegoro under teak forest and opened area by using completely randomised design (2x5) first factor was location and second factor was fertilizer level (LEISA model). The result showed that first factor affected DM production ( $P < 0.01$ ). The coleration between both factors also affected DM production ( $P < 0.01$ ). *Indigofera z.* Showed best result of planting using (50% organic + 50% anorganic) fertilizer on under teak forest and (75% organic and 25% anorganic) fertilizer on opened area ( $P < 0.01$ ). *Pueraria javanica* showed best result of planting using (100% anorganic) fertilizer on under teak forest and (50% organic + 50% anorganic) fertilizer on opened area ( $P < 0.01$ ). This study conclude that *Indigofera zoolingeriana* and *Pueraria Javanica* have a good adaptability in dry land areas and have capability to integrated with teak forest, it means *Indigofera zoolingeriana* as tree legume and *Pueraria javanica* as bush legume potential to developed on bojonegoro. Keywords: drought stress, fertilizer, *Indigofera zoolingeriana*, Leguminose, *Pueraria javanica*

## Introduction

Forage is a ruminant's husbandry important factor to boots Indonesia's husbandry development. Leguminose is a kind of feed crop that has better quality of protein than tropical grass, beside legume also tend to produce more digestible dry matter per hectare (Odihiambo and Bomke, 2001).

The limited availability of agricultural land due to the usage of land for habitation causing the importance of the dry land should be optimized as forage development area, especially for legume. Bojonegoro is one of district located in East Java Province. Bojonegoro has enough area to develop as forage centre, but it has dryland condition, high light intensity and limited water availability in dry season. Those problem should be solve by integrate legume with teak forest and determine the dose of optimal fertilizer to sustain green forage growth and produce good nutritional quality .

*Indigofera zoolingeriana* is a kind of tree legume which has high nutrition and high production with drought tolerant characteristic (Hassen *et al.*, 2007). In other side, *Pueraria javanica* is a kind of herbs legume which has high quality of protein and the production reach to 10 tones per hectare (Valentim & Andrade, 2005). *Pueraria javanica* has low light intensity tolerant characteristic. Because of that, both of legume, has potency to be developed on dry land by integrated system.

This study aim to learn the morphology and nutrition response of *Indigofera zoolingeriana* and *Pueraria javanica* in integrated system with teak forest on dry land in Bojonegoro and to determine the optimal fertilizer dose to get good legume quantity and quality.

## Materials and Methods

This study consisted of two phases, the first phase was done in the greenhouse of Agrostology Unit in farm field laboratory, Bogor Agricultural University (IPB) on October to December 2014. The second phase was done in the Sambeng Village, District Kasiman, Bojonegoro on December 2014 to January 2015.

The first phase used sixteen seeds for each kind of legume (*Indigofera zoolingiana* and *Pueraria javanica*) that planted in medium contained organic and inorganic fertilizers (Urea, SP36, KCL). This phase used Completed Random Design (CRD) with four treatments and four replications. Those treatments were: P1: 100% field water capacity, P2: 75 % field water capacity, P3: 50 % field water capacity, P4: 25 % field water capacity. Watering was done every six days according to the treatment dose to maintain water content on growing media. The parameters were: plant morphology such as plant height, diameter of stems and amount of leaves.

The experimental design in the second phase of this study used Completed Random Design (CRD) with five treatments and four replications. Each replication consisted of eight plants. The amount of each type of plants used 160 seeds. Those five treatments were: T1: 100% organic fertilizer, T2: 75 % organic fertilizer + 25% chemical fertilizer, T3: 50% organic fertilizer + 50 % chemical fertilizer. T4: 25 % organic fertilizer + 75% chemical fertilizer. T5: 100 % chemical fertilizer.

The parameters were: plant morphology i.e.: plant height and DM production. Nutrition Parameter plant is proximate plant using method AOAC (2005) and Van Soest (1991).

## Results and Discussion

The first phase of this study showed that a response of plant on drought stress based on morphological aspects in Table 1.

Table 1. *Indigofera zoolingiana* and *Pueraria javanica*'s response based on morphological aspect of plants

Variables measured	<i>Indigofera zoolingiana</i>				<i>Pueraria javanica</i>			
	P1	P2	P3	P4	P1	P2	P3	P4
Plant's height (cm)	52.25±11.4a	65.42±12.8ab	75.55±12.8b	75.37±10.2b	87.67±21.6a	121.85±22.3ab	124.57±34.8ab	148.20±11.9b
Stem diameter (cm)	0.35±0.03a	0.41±0.04ab	0.52±0.07c	0.48±0.06bc	0.35±0.08	0.34±0.08	0.36±0.06	0.36±0.04
Amount of leaves	11.75±1.5a	17.25±5.1b	20.25±5.5b	18.25±1.0b	11.00±3.8a	17.75±5.5ab	22.75±1.25b	24.75±5.7b

Description: P1: the moisture content of 25% field capacity; P2: 50% field capacity; P3: 75% field capacity; P4: 100% field capacity. Figures followed by different letters in the same row showed there is a significant effect ( $P < 0.05$ ).

Results showed that drought stress treatment has significant effect ( $P < 0.05$ ) on *Indigofera zoolingiana* plant's height, stem diameter and amount of leaves. The highest morphology response values of *Indigofera zoolingiana* were showed by treatment P3 and P4, while the lowest value was showed by treatment P1. This was due too P3 and P4 were optimal conditions for plant growth while on P1 is severe drought stress conditions that can inhibit plant growth rate. Prasad *et al.* (2008) state that generally the division and growth of plant cells is strongly influenced by the presence of water both inside and outside the plant cell, because water is a medium growth and cell metabolism. The decrease percent age in plant height, stem diameter, and amount of leaves of P4 (100% field capacity) to P1 (25% field capacity) respectively were 30.8%, 32.6%, 41.9%.

Results showed that drought stress treatment has significant effect ( $P < 0.05$ ) on *Pueraria javanica* plant height, stem diameter and amount of leaves. Plant height and amount of leaves showed that the best values was treatment P4 while the lowest value was P1. Treatment P4 was an optimal condition for plant's growth while on P1 was severe drought stress conditions that can inhibit plant growth rate. The decrease percentage of plant height and amount of leaves on treatment P4 to P1 were 40.9% and 55.5%, respectively.

Table 2 showed that response of *Indigofera zoolingiana* in integrated with teak forest was significant ( $P < 0.05$ ) on fertilizer treatment.

Table 2. Morphology observation result on *Indigofer zoolingiana* in Bojonegoro

Variables measured	<i>Indigofera zoolingiana</i>									
	N1	N2	N3	N4	N5	T1	T2	T3	T4	T5
Plant height	81.7	71.6	93.2	95.2	73.5	99.4	93.2	66.5	59.7	56.9
Biomass production (g)	74.0ab	30.0a	88.8b	24.8a	56.5ab	38.2a	50.7ab	71.5b	144.4c	44.0a

Description: N : Planting location in the shade of teak; T : Planting location without shade; 1,2,3,4,5 which follows the description of the location of planting (N or T) is a description of fertilizer treatment; Numbers followed by different letters in the same row showed there is significant effect ( $P < 0.01$ ).

The highest DM value was showed by fertilizer treatment N3 (50% organic fertilizer + 50% anorganic) that is 88.8 gram and plant height 93.2 cm. *Indigofera zoolingeriana* that planted on opened area showed the highest response at treatment T4 (25% organic fertilizer + 75% anorganic) i.e.: DM value was 144.4 gram and plant height was 59.7cm.

Table 3. Morphology observation result on *Pueraria javanica* in Bojonegoro

Variables measured	<i>Pueraria javanica</i>									
	N1	N2	N3	N4	N5	T1	T2	T3	T4	T5
Plant height	72.7	52.5	92.00	81.2	57.50	97.13	88.00	122.2	74.50	130.2
Fresh weight harvest (g)	83.2b	42.7a	100.9c	65.5ab	112.0c	187.3b	187.2b	184.8b	136.8a	128.4a

Description: N: Planting location in the shade of teak; T: Planting location without shade; 1,2,3,4,5 which follows the description of the location of planting (N or T) is a description of fertilizer treatment; Numbers followed by different letters in the same row showed there is significant effect (P<0,01).

Response of *Pueraria javanica* that plant integrated with teak forest (Table 3.) showed significant effect (P<0.05) on fertilizer treatment. The highest response showed in fertilizer treatment N5 (100% anorganic fertilizer) that was DM value 112.0 gram and plant height 57.50 cm. *Pueraria javanica* that planted on opened area showed the highest response in treatment T3 (50% organic fertilizer + 50% anorganic) that was DM value 184.8 gram and Plant height 122.2cm.

Table 4. Proximate and Van Soest result on plants in di Bojonegoro

Variables measured	<i>Indigofera zoolingerian</i>		<i>Pueraria Javanica</i>	
	N3	T4	N5	T3
Proximate				
Water Content (%)	90.97 ± 0.63	90.32 ± 0.43	93.17 ± 0.16	93.12 ± 0.65
CP (%)	20.08 ± 0.04	21.41 ± 0.13	9.51 ± 0.31	11.78 ± 0.33
CF (%)	17.45 ± 0.98	17.55 ± 0.73	23.94 ± 2.94	37.29 ± 0.42
Lipids (%)	1.30 ± 0.04	2.26 ± 0.07	0.61 ± 0.01	0.78 ± 0.07
Ash (%)	16.29 ± 0.95	15.00 ± 0.66	27.20 ± 3.20	11.36 ± 1.23
Van Soest				
ADF	34.20 ± 1.97	35.87 ± 1.40	57.70 ± 3.52	46.30 ± 3.27
NDF	35.86 ± 0.07	39.08 ± 1.64	55.34 ± 0.80	52.89 ± 1.84
Lignin	16.65 ± 3.62	8.81 ± 2.39	19.80 ± 1.78	26.90 ± 0.35
Selulosa	16.99 ± 3.39	27.06 ± 1.90	37.90 ± 5.28	19.41 ± 2.93

Description: Analytical result from LIPI Laboratory (2015).N: Planting location in the shade of teak; T: Planting location without shade; CP: Crude Protein; CF: Crude Fiber; 1,2,3,4,5 which follows the description of the location of planting(N or T) is a description of fertilizer treatment.

Nutrient and fiber fraction of *Indigofera zoolingeriana* and *Pueraria javanica* were showed in Table 4. Proximate and Van Soes analyses (CP, CF, Lipid and Ash) on *Indigofera zoolingeriana* and *Pueraria javanica* plants showed that has no differences between plants grown in opened area with those that integrated with teak forest.

## Conclusion

Legume tree *Indigofera zoolingeriana* and legume grass *Pueraria javanica* has potential to be developed on dry land because both of them has has high adaptability to drought stress. Integration between legume with teak forest were optimal at fertilizer dose 50% organic fertilizer +50% anorganic for *Indigofera zoolingeriana* and 100% anorganic fertilizer for *Pueraria javanica*. On opened area, plant were optimal at fertilizer dose 25% organic fertilizer +75% anorganic for *Indigofera zoolingeriana* and 50% organic fertilizer +50% anorganic for *Pueraria javanica*.

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# The Diversity and Quality of Forages Used for Feeding of Goat in Payakumbuh of West Sumatra

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## Abstract

*The present research was aimed to evaluate the diversity and quality of forages used for feeding of dairy goats in Payakumbuh regions of West Sumatra. Sample of forages were taken in 3 different sampling times from 8 goat farms. The fresh samples from 5 different sampling points at each farm were weighed and then sorted by plant species for identification of botanical composition. Seven highest percentages of individual plant and a mixture of other plants were chopped, dried and ground for chemical analysis. Parameter measured included botanical composition, DM and nutrient content of CP, CF, crude ash and minerals of Ca, P, Fe, Cu, Mn and Zn. Results showed that there were about 47 kinds of vegetation used for feeding for goats in Payakumbuh. They were composed of 40% of native grasses, 34% of broadleaves, 19% of legumes, and 6% tree leaves, respectively. The seven highest portion of forages was *Axonopus sp*, followed by *Panicum sp*, ferns, *Centro sp*, gamal and cassava leaves. Legumes and tree leaves were found containing lower CF and higher CP and essential minerals of Ca, P, Fe, Cu, Mn and Zn in compare to grasses and broadleaves. It was concluded that there were diversity forages resources available for feeding of goats in Payakumbuh regions, but the nutritious green fodders of legumes and tree leaves were still used in limited portion.*

**Keywords:** *dairy goat nutrition, minerals, wild forages*

## Introduction

Payakumbuh which cover Payakumbuh city and 50 Kota district is one of the areas of potential for development of goat farming in West Sumatra. Goat farms are dominated by small-scale enterprises which the major breeds are the Kacang and Etawah grade (PE) goats. They are distributed mainly in six sub districts of Lareh Sago Halaban, Harau, Mungka, Luhak, East Payakumbuh and West Payakumbuh. Khalil and Reswati (2014) reported that raising dairy goat has better prospect in compare to meat-type due to potential market for goat's milk and higher price of bucks in Payakumbuh. The average flock size of about 35.9 of dairy goat/farm was much higher in compare to meat-type breeds of 14.3 goats/farm (Khalil and Reswati, 2014).

Dairy goats are raised intensively with cut and carry feeding system. The feeding is based primarily on the use of native vegetation, tree leaves and agricultural by-products. The wild vegetation forages are derived from diverse sources, like plantation areas, river banks, rice fields, forest edges and road sides. Vegetation grown in such areas are considered as weeds, not treated and comprise of various types of wild plants, such as native grass, legumes, broadleaf species and ferns. These feed were not only vary in nutrient content, but also are often of poor mineral and DM (Khalil *et al.*, 2014). Thus, determining the botanical composition and nutrient content of dominant species may be useful for improvement of feeding strategy.

The present research was aimed to evaluate forage resources that are available in terms of the species, botanical composition and quality used for feeding of goats by small-scale enterprises in Payakumbuh regions of West Sumatra.

## Materials and Methods

The study was initiated by collecting samples of forages from 8 goat farms located in Payakumbuh city and 50 Kota district. The farms have about 24 goats/farm in average. They were distributed in 7 different sub districts of East Payakumbuh, West Payakumbuh, Lareh Sago Halaban, Luhak, Arau, Akabiluru and Tanjung Aro. These areas are dominated by annually small-scale crop estates as potential sources of green fodder.

Samples of feed in fresh form were taken in 3 different sampling times from 5 different sampling points at each farm. The fresh samples were weighed and then sorted by plant species for identification of botanical composition. Seven highest percentages and most frequent species were selected and chopped. Representative samples of about 100-150 g were dried in a forced draught oven at 60 °C for 24 hours and ground in meal form prior to analysis for dry matter (DM), crude ash, crude protein (CP), crude fiber (CF) and minerals of Ca and P.

DM and nutrient contents of crude ash, CP and CF were determined using the procedure described by AOAC (2005). The concentration of minerals was determined using an atomic absorption spectrophotometer (AAS, 1980). All analysis results were reported on DM basis. Parameter measured included botanical composition, DM and nutrient content of CP, CF, crude ash and minerals of Ca and P. Data on nutrient and mineral content were subjected to analysis of variance (ANOVA) in completely random design of 7x3 consisting of 7 forage species and 3 replicates. Duncan's Multiple Range (DMRT) was applied to separate means. Differences were considered significant at  $P < 0.05$  (Steel *et al.*, 1997).

## Results and Discussion

Results showed that there were about 47 plant species used for feeding of goats in the study sites. They were composed of about 40% of grasses, 34% of broadleaves, 19% of legumes and 6% tree leaves, respectively. It shows that there were diversity kinds of fodder available for feeding of goats in Payakumbuh. Goats have the unique ability and the tendency to utilize wide diversity of plants species such as woody plant species, forbs and grasses which are not generally consumed by other domestic livestock (Hart, 2001). Therefore, goats were favorable alternative and suitable incorporated for small-scale farmer or part-time livestock producer in Payakumbuh. This data also shows that farmers in Payakumbuh were able to explore the potential of various forage sources for feeding of their goats.

Grasses were dominated by native species of *Axonopus compressus*, *Panicum repens*, *Paspalum conjugatum*, *Ottochloa nodosa*, *Brachiraria milliformis*, *Brachiaria mutica*, *Ischaenum mucunoides*. *Axonopus sp* which composed of about 23% was found the most important grass species, followed by *Panicum sp* (5.3%) and *Paspalum sp* of 4.0%. *Axonopus* and *Paspalum* which are known as high palatable and shade tolerant species was commonly found growing as weeds under rubber and palm crop plantations (Wong, 1990), while *Panicum* was widely found in the area of banana, and coconut plantations. Sub district of Lareh Sago Halaban, Harau, Mungka, Luhak, East Payakumbuh and West Payakumbuh are dominated by annually small-scale crop estates of cacao, coconut and banana.

The two most important legumes were *Centrosema pubescens* and *Desmodium sp*. Broadleaves were dominated by *Amaranthus sp*, *Borreria alata* and *Miknia cordata*. There were two potential leaves available in Payakumbuh, i.e. gliricidia and cassava leaves. *Gliricidia sepiumis* commonly used as live fences, while cassava is widely planted to produce cassava roots for making snack foods. In Nigeria, important plant families that contribute to the most preferred forage resource base of goats include tree and shrub legumes, which formed 15.80% of which families Fabaceae (Papilionaceae), Caesalpinadeae and Mimosaceae comprised 2.63%, 2.63% and 10.53% respectively (Obua, 2014).

Table 1 shows DM, crude nutrient and mineral content of seven dominant plant species fed for goats in Payakumbuh. The nutritive values of dominant plant species used for feeding goat in Payakumbuh were found relatively high. The crude protein content of native grasses ranges from 11.3 to 13.1 %, while legumes from 17-23%. Legumes and tree leaves were found containing lower CF and higher CP in compare to grasses. Legumes contained also relatively high P of about 11-13 g/kg DM. The content of crude protein and minerals of Ca and P of fern was found equal to native grasses. The highest CP of about 28-27% and lowest CF content (13-21%) was shown by tree foliage of gliricidia and cassava leaf. Fodder leaves were also found as good mineral Ca sources with significantly highest Ca content of about 15-20 g/kg DM. Tree foliage which composed of young leaves, stalks, seeds and floral parts of the vegetation components have a high nutritional value (Handayanta *et al.*, 2014). Ajayi *et al.* (2005) reported that gliricidia foliage was a good protein sources for goats and had a crude potent content of 29.3%. High concentration of Ca in cassava leaf has also been reported by Fasuyi (2005). Adiwimarta *et al.* (2014) reported that cassava leaf was not only a good protein sources but also had an anthelmintic effect for goats.

Table 1. Crude nutrient and mineral content of forages fed for goat in Payakumbuh

Plant species	DM (%)	CP (%)	CF (%)	Ca (g/kg)	P (g/kg)
Axonopus	23.9±0.2 <sup>ab</sup>	11.3±0.3 <sup>c</sup>	35.3±3.5 <sup>a</sup>	2.1±0.3 <sup>d</sup>	8.1±0.7
Panicum	22.7±0.6 <sup>ab</sup>	13.1±0.2 <sup>c</sup>	37.4±1.9 <sup>a</sup>	2.2±0.1 <sup>d</sup>	5.8±1.1
Centrosema	17.1±0.2 <sup>b</sup>	17.2±1.4 <sup>b</sup>	29.2±2.6 <sup>b</sup>	3.5±0.5 <sup>d</sup>	12.6±0.5
Desmodium	14.6±0.1 <sup>b</sup>	23.2±0.3 <sup>a</sup>	25.6±3.2 <sup>b</sup>	8.2±1.7 <sup>c</sup>	11.3±2.4
Gliricidia leaves	24.9±0.2 <sup>ab</sup>	26.8±1.0 <sup>a</sup>	12.7±1.2 <sup>d</sup>	20.1±1.9 <sup>a</sup>	7.4±0.9
Cassava leaves	28.5±1.0 <sup>a</sup>	25.3±1.7 <sup>a</sup>	21.1±1.6 <sup>c</sup>	15.0±0.4 <sup>b</sup>	11.4±0.9
Fern	27.2±0.6 <sup>a</sup>	11.2±0.6 <sup>c</sup>	28.7±0.5 <sup>b</sup>	2.8±0.3 <sup>d</sup>	7.2±1.9

Different superscript in the same line means significantly different (P<0.05)

Even though native grasses have slightly lower crude protein and mineral and higher CF content, their combinations with legumes and tree leaves which contain high crude proteins and minerals make them good forages. Due to their high nutritional content, the use of fodder from tree leaves should be increased in the feeding of goats. Because of relatively good fodder feeds, the average milk production of about 0.8 l/head/day in Payakumbuh (Kurnia *et al.*, 2015) was comparable the results reported by Novita *et al.* (2006), where lactation milk yield was 0.5-0.9 liters per day.

However, farmers should give attention on the daily quantity of forages offered to meet DM and nutrient requirement of their livestock due to relatively low DM content of the forages of about 22%. The average DM content of legumes of 14-17% was significantly lower in compare to grasses (23-24%) and tree leaves (25-29%).

## Conclusion

It was concluded that there is wide variety of plant species utilized as green fodder feed for goats in Payakumbuh. The forages offered mainly composed of native grasses, broadleaves, legumes and tree leaves. Legumes and tree leaves generally contained higher CP and minerals in compare to grasses and fern. Due to low in dry matter content of the most dominant species, it was therefore necessary to take into account the quantity of fresh forages for feeding goats under confinement to meet their nutrient and dry matter needs.

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# The Addition of *Arbuscular mycorrhizal* Fungi in Enhancing Productivity and Drought Tolerance Mechanisms of *Indigofera zollingeriana*

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## Abstract

*Arbuscular mycorrhizal fungi (AMF) is symbiotic associations between plant roots and fungi. The main role of the AMF is to increase nutrient and water uptake by the host plant. The purpose of this research is to study the role of AMF in enhancing productivity and tolerance mechanisms of Indigofera zollingeriana in drought conditions. This study used a completely randomized design with four treatments: M0W0 (without AMF), M0W1 (without AMF with drought), M1W0 (with AMF) and M1W1 (with FMA and drought). The parameters measured were leaf water potential, leaf water content, shoot and root dry weight, proline, soluble sugars. Data were tested using Analysis of Variance (ANOVA) and the differences among treatments' means were examined by Duncan Multiple Range Test. The results showed that plants inoculated AMF with and without drought increased shoot and root dry weight, but decreased proline and soluble sugars ( $P < 0.01$ ). Drought can reduce leaf water potential, shoot and root dry weight, but increase in proline, soluble sugars and productivity in plant with AMF ( $P < 0.01$ ). Mechanisms of drought tolerance through accumulation proline and soluble sugars on Indigofera zollingeriana.*

*Keywords: Arbuscular mycorrhizal fungi, drought, Indigofera zollingeriana, proline, soluble sugars.*

## Introduction

*Arbuscular mycorrhizal fungi (AMF) is the symbiotic relationship between plant roots and fungi, it play a role in improving the absorption of nutrients and water by the host plant. Drought stress is one of the environmental factors as a factor limiting the growth, development and yield. Plants can respond and adapt to drought stress by altering the metabolism of the cellular level and apply a variety of defense mechanisms (Smith and Read, 2008). In the drought conditions, mycorrhizal plants and without AMF will organize different expression of several genes related stress in root tissue (Ruiz-Lozano *et al.*, 2006). The Contributions of symbiosis AMF on drought tolerance, resulting from the combined effects of physical, nutritional, physiological, and cellular effects (Ruiz-Lozano, 2003). The aim of this research was to study the role of AMF in enhancing productivity and tolerance mechanisms of Indigofera zollingeriana in drought conditions.*

## Materials and Methods

The soil samples is from around the cage of the Faculty of Animal Husbandry, manure, mycorrhizal, NPK fertilizer and chemicals for analysis of proline, water soluble carbohydrate (WSC), and others. The tools used in this study is a shovel, scales capacity of 5 kg, plastic, pot capacity of 5 kg, shears, digital scales, rulers, plastic mulch, WP4 meter, oven, refrigerator, freezer, paper /envelopes, hot plate, ependaf, vortex, centrifuge, spectrometers, desiccator, ice gel, coolbox, waterbath and others. This study uses a completely randomized design with four treatments: M0W0 (without AMF), M0W1 (without AMF with drought), M1W0 (with AMF) and M1W1 (with AMF with drought). The parameters measured were leaf water potential, water content shoot, root and shoot dry weight production and, proline content (Bates, 1973) and soluble sugars (modification by Buysse & Merckx 1993). Data were tested using Analysis of Variance (ANOVA) and the differences among treatments' means were examined by Duncan Multiple Range Test (Steel and Torrie, 1995). Data was processed by using Excel and SAS programme.

## Results and Discussion

The response of AMF additions and drought stress on water potential can be seen in Figure 1. Plants that are not experiencing drought stress shows water potential between -1.28 to -2.05. Plants that experienced drought stress shows the potential drop of water on 20<sup>th</sup> days to -5.98 and -9.91. Plants that are added to the AMF on normal or drought conditions shows water potential decreased. Lower water potential drought conditions (M1W1) indicates the plant is trying to still get water, so that growth is not hampered due to lack of water. Shoot water content in the treatment W1M1 not decreased on 20<sup>th</sup> days, but the treatment MoW1 decreased (Table 1). Querejeta *et al.* (2003, 2006) reported that in field studies mycorrhizal relationship with water, mycorrhizae can improve the water plant. Water potential in plants between -2 to -3.5 MPa, while water potential in the root zone is between -1.5 and -2 MPa.

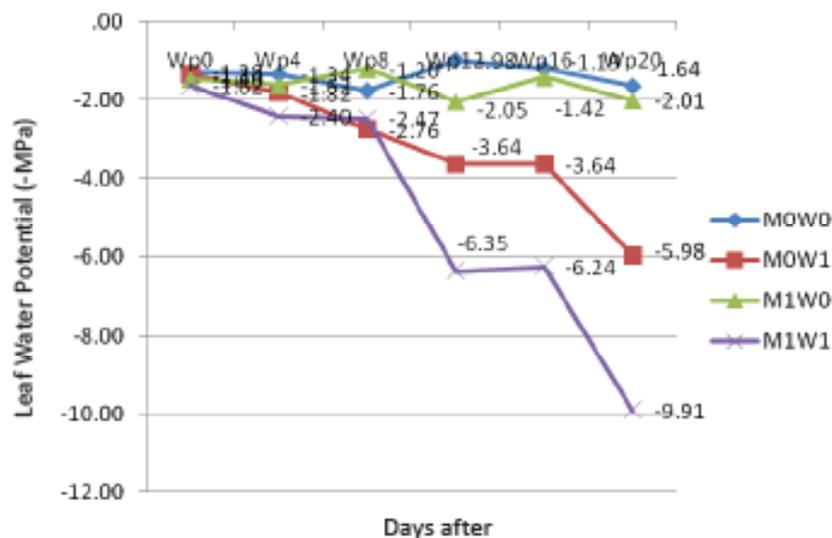


Figure 1. The addition of FMA and drought on leaf water potential

The AMF additions response and drought stress on the production of roots and shoot can be seen in Figure 2. Drought causes decreased production of dried roots, especially in non-mycorrhizal plants. The Addition of AMF shows increased production of dried roots both in normal and drought conditions. The same pattern occurs in the production of dried shoot. Karti (2004) reported that the interaction between the AMF and drought stress were not significantly different. Growth and crop production has decreased in plants without the AMF and FA plants with better production. Karti *et al* (2012) reported that plants with AMF on drought stress is higher productivity compared to plants without AMF.

Table 1. The addition of FMA and drought on water content

Perlakuan	Shoot Water Content (%)					
	0	4	8	12	16	20
MOW0	89.52	91.07	87.72	86.35	91.04	88.48
MOW1	89.04	90.13	86.61	86.98	88.62	87.79
M1W0	89.71	90.17	90.08	87.76	90.38	88.92
M1W1	87.13	91.98	85.73	87.23	87.31	87.38

The response of AMF additions and drought stress on proline production can be seen in Figure 3. Plants on drought stress with and without AMF increased proline content (MOW1 and M1W1). The greatest increase at 16<sup>th</sup> day occurred in plant with AMF (M1W1). Proline is needed for improvement osmotic adjustment through decreasing the water potential, so that the plant can still absorb water in drought stress conditions. In the non-mycorrhizal plants proline content is lower, so the potential water generated is greater than the mycorrhizal plants in drought conditions. Water stress significantly increases the accumulation of proline in plants (Verslues & Sharman, 2010), corn (Khani & Heidari, 2008). This is possible because proline play an important role in preventing or reducing the harm caused by lack of water (Khani & Heidari, 2008).

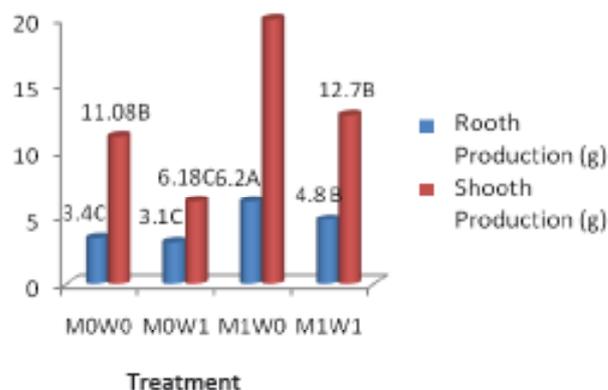


Figure 2. The addition of AMF and drought stress on shoot and root production

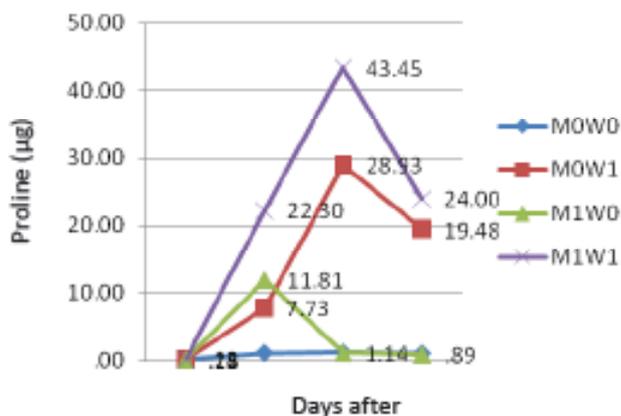


Figure 3. The addition of AMF and drought stress on proline content

The response of AMF additions and drought stress on soluble sugars content, can be seen in Figure 4. The plants without mycorrhizal on drought stress (MoW1) increased the soluble sugar, whereas in mycorrhizal plants (M1W1) is lowest. In the drought stress conditions the plants with mycorrhizal (M1W1) do not require soluble sugars for osmotic adjustment, due to the increasing of proline and the proline was higher when compared with non-mycorrhizal plants (MoW1). The increasing soluble sugars in plants without mycorrhizal can lower water potential. Wu *et al.* (2007), plants with AMF accumulate soluble sugar, starch dissolved and total non-structural carbohydrates higher in the leaves and roots of the plant without the AMF. The lower accumulation of soluble sugars showed that the plant managed to avoid drought and do not requires osmotic adjustment so as not to damage (Song, 2005).

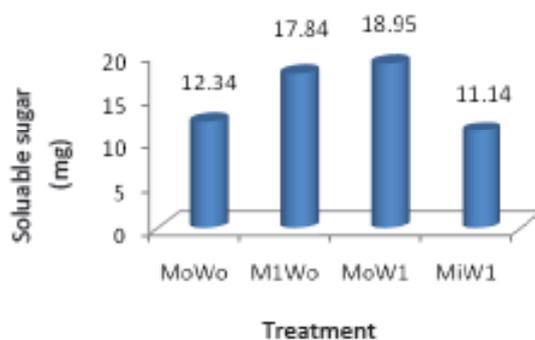


Figure 4. The addition of AMF and drought stress on soluble sugar

## Conclusion

Inoculation AMF without drought can increase productivity. Drought can reduce leaf water potential and productivity, but plants with AMF increased proline and productivity. Mechanisms of drought tolerance in *Indigofera zollingeriana* through the accumulation of proline for mycorrhizal plant and soluble sugar for non-mycorrhizal plant.

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# Growth and Productivity of Different Sorghum Varieties Cultivated with *Indigofera* in Intercropping System

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## Abstract

The experiment is aimed at recognizing growth and production of different varieties of sorghum that was grown with high valuable legume *indigofera* in intercropping system and to determine the suitable variety of sorghum that produced high grain and forages in the system. The experiment was done at UP3J Jonggol, from November 2014 to March 2015. This experiment was conducted using the Completely Randomized Design with two factors and four replications. The First factor was sorghum varieties (patir 3.2 (S1), patir 3.7 (S2), and citayam (S3)). The second factor was *indigofera* composition (0 % *indigofera* (I0), 30 % *indigofera* (I1), 40 % *indigofera* (I2), and 50 % *indigofera* (I3)). Datas were analyzed using analysis of variance and HSD test. The results showed that sorghum variety citayam had fastest ( $P < 0.01$ ) growth indicated by plant height, and highest ( $P < 0.01$ ) productivity indicated by leaf weight and stem weight. The results also revealed that combination of sorghum varieties and *indigofera* in an intercropping system led to an increase of total above ground biomass yield of experimental plots. The suitable variety of sorghum and *indigofera* was found in plots cultivated combination of patir 3.2 with 50 % *indigofera*, patir 3.7 with 50 % *indigofera*, and citayam with 40 % *indigofera*. Highest biomass yield was found in plots cultivated combination patir 3.7 with 50 % *indigofera*.

Keywords: growth, *indigofera*, intercropping, productivity, sorghum

## Introduction

Intercropping is one of the most common cultivation practices used in sustainable agricultural system, which plays important role in increasing land productivity and yield stability. This cropping system improves resource utilization and environmental factors (Najafi and Kestehgar, 2014). The main purpose of intercropping is to produce a greater yield on a given piece of land by making use of resources that would otherwise not be utilized by a monocropping efficiently (Moradi *et al.*, 2014). The main advantage of intercropping is the more efficient utilization of the available resources and the increase productivity compared with each sole crop of the mixture. Intercropping can conserve soil water by providing shade, reducing wind speed and increasing infiltration with mulch layers and improved soil structure. Enhanced productivity of multispecies agroecosystems (intercropping) compared with that of monospecific agro ecosystems (each of the component species being grown alone) may be explained by two major processes that result in improved resource use: complementarity and facilitation.

In the intercropping system root interaction could increase the root activity and microbial quantity in the rhizosphere (Zhang, 2013). Rhizospheric interspecies interaction may also affect nutrient availability and uptake in intercropping. Alley cropping can maintain and sequester soil C and N beyond organic conservation tillage and more than conventionally tilled, chemically fertilized treatment (Jacobsen, 2008).

Sorghum is one of important cereal crops grown in the rainfed areas over an area of 42 million ha for grain and fodder purpose in the semi-arid tropic of Africa, Asia and Latin America (Reddy *et al.*, 2004). Sorghum has a high yield potential, comparable to rice, wheat, and maize. On a field basis, yields have exceeded 11 ton ha<sup>-1</sup>, with above average yields ranging from 7-9 ton ha<sup>-1</sup> where water is not limited. In the areas where sorghum is commonly grown, yields of 3-4 ton ha<sup>-1</sup> are obtained under normal condition (House, 1985). Sorghum is also known to have wide adaptability, ranging from lowland, medium to highland altitude. As an alternative animal feed sorghum have good nutritional content, short-lived (100-110 days), resistant to drought and resistant to pest attack. Sorghum also has great potential to supplement fodder resources because of its wide adaptation, rapid growth, high green and dry fodder yields with high ratoonability (Reddy, 2004).

Indigofera sp. is a plant of the legume group (family Fabaceae) with genus Indigofera and has 700 species spread in Africa, Asia, Australia, and North America (Schrire *et al.*, 2009). Indigofera species have great promise as forages for ruminants. It is a potential legume because it has a good growth with high production and nutritive value (Hassen *et al.*, 2008). Their high protein levels and ability to tolerate drought, floods and salinity make them agronomically desirable, while their deep-rooted growth form, ability to respond to small rainfall events and resistance to herbivory make them potentially valuable cover crops and forage species for semi-arid and arid areas.

There are no reports on growth and production of different varieties of sorghum that was grown with high valuable legume Indigofera in intercropping system. This study was conducted to test the growth and productivity of intercropping suitable composition different varieties of sorghum that was grown with high valuable legume Indigofera.

## Materials and Methods

The experiment was done at UP3J Jonggol, from November 2014 to March 2015. This experiment was conducted using Completely Randomized Design with two factors and four replications. The first factor was sorghum varieties (patir 3.2 (S1), patir 3.7 (S2), and citayam (S3)). The second factor was indigofera composition (0 % indigofera (I0), 30 % indigofera (I1), 40 % indigofera (I2), and 50 % indigofera (I3)). Data were analyzed using analysis of variance and HSD test.

Plant density of sorghum-indigofera in plots was done by calculating the ratio of greenery needs to be provided by each plant with a production capacity per plant in a land area (m<sup>2</sup>). After considering sorghum plant space at 70 cm x 20 cm x 40 cm in cropping alley along indigofera at a 120cm between rows, plots area were then resulted at 10m x 10m with fixed number of sorghum plants were as many as 360 individual plants. Based on that calculation, composition of 70% sorghum and 30% indigofera was for sorghum and indigofera 360 and 48 individual plants respectively; for composition of 60 % sorghum and 40% indigofera were sorghum 360 plants and indigofera 78 plants; and 50% sorghum and 50% indigofera were sorghum 360 plants and indigofera 114 plants.

A manual cutting was conducted when the sorghum reach 80 % flowering. Growth parameters including plant height (cm), number of leaf (blad), width of leaf (cm), and length of leaf (cm) were recorded; and productivity including leaf weight (gr), stem weight (gr), and panicle weight (gr), and biomass (ton/ha) were recorded as well. Plant biomass was measured using total heavy plant of sorghum and indigofera consisting of leaves, stems and panicles (sorghum).

## Results and Discussion

The plant height, number of leaf, width of leaf, and length of leaf are shown in Table 1. There were significant different growth characters ( $P < 0.01$ ) among sorghum cultivars. There were non-significant differences for composition of indigofera and interaction between varieties of sorghum and indigofera composition for all variables. Any changes in the composition of indigofera had no effect on growth of sorghum varieties.

The results of productivity indicated by leaf weight, stem weight, and panicle weight (Table 2) of sorghum variety cultivated with indigofera in intercropping system. There were significant differences ( $P < 0.05$ ) between sorghum cultivars in all growth characters. There were significant differences ( $P < 0.05$ ) between indigofera composition for stem weight but non-significant differences for leaf weight and panicle weight. There were no interactions between varieties of sorghum and indigofera composition for all variables.

The results of productivity indicated by biomass (Table 3) of sorghum variety cultivated with indigofera in intercropping system. There were significant differences ( $P < 0.05$ ) between sorghum cultivars in biomass yield, but nonsignificant for indigofera composition and interaction between varieties of sorghum and indigofera composition. Although non-significant interaction between varieties of sorghum and indigofera composition, but the data show that high product biomass yield for suitable composition different varieties of sorghum that was grown with high valuable legume indigofera were patir 3.2 and patir 3.7 with 50 % indigofera, and citayam with 40 % indigofera.

Table 1. Plant height (cm), number (blade), width (cm), and length (cm) of leaf.

Treatment	Sorghum			
	Plant height	Number of leaf	Width leaf	Length leaf
Sorghum				
S <sub>1</sub>	209.03 ± 7.34 <sup>a</sup>	6.33 ± 0.64 <sup>a</sup>	8.27 ± 0.39	97.53 ± 2.80 <sup>a</sup>
S <sub>2</sub>	215.84 ± 9.81 <sup>a</sup>	6.95 ± 1.30 <sup>a</sup>	8.39 ± 0.58	95.52 ± 8.85 <sup>a</sup>
S <sub>3</sub>	274.28 ± 14.54 <sup>b</sup>	8.43 ± 1.20 <sup>b</sup>	8.73 ± 0.79	107.76 ± 9.28 <sup>b</sup>
Indigofera				
I <sub>0</sub>	229.15 ± 28.80	7.05 ± 1.48	8.84 ± 0.61	103.43 ± 7.81
I <sub>1</sub>	230.07 ± 30.51	7.41 ± 1.33	8.38 ± 0.61	100.92 ± 7.21
I <sub>2</sub>	234.11 ± 37.60	7.20 ± 1.30	8.41 ± 0.72	99.13 ± 10.34
I <sub>3</sub>	238.87 ± 31.57	7.28 ± 1.58	8.23 ± 0.44	97.58 ± 10.94
S vs I				
S <sub>1</sub> *I <sub>0</sub>	206.95 ± 8.01	6.25 ± 0.68	8.44 ± 0.43	97.55 ± 4.46
S <sub>1</sub> *I <sub>1</sub>	208.99 ± 4.09	6.60 ± 0.85	8.47 ± 0.24	98.55 ± 3.00
S <sub>1</sub> *I <sub>2</sub>	206.55 ± 2.58	6.00 ± 0.69	8.31 ± 0.27	96.70 ± 1.86
S <sub>1</sub> *I <sub>3</sub>	213.65 ± 1.81	6.45 ± 0.30	7.87 ± 0.36	97.30 ± 2.07
S <sub>2</sub> *I <sub>0</sub>	213.98 ± 8.79	6.35 ± 1.48	8.68 ± 0.45	99.75 ± 1.53
S <sub>2</sub> *I <sub>1</sub>	214.45 ± 9.13	7.25 ± 1.59	8.18 ± 0.56	98.83 ± 0.69
S <sub>2</sub> *I <sub>2</sub>	211.68 ± 11.55	7.55 ± 1.00	8.26 ± 0.84	91.65 ± 12.68
S <sub>2</sub> *I <sub>3</sub>	223.25 ± 9.16	6.65 ± 1.25	8.45 ± 0.50	91.85 ± 12.32
S <sub>3</sub> *I <sub>0</sub>	266.53 ± 8.52	8.55 ± 0.91	9.41 ± 0.53	113.00 ± 3.87
S <sub>3</sub> *I <sub>1</sub>	266.78 ± 24.47	8.38 ± 1.06	8.48 ± 0.96	105.38 ± 11.89
S <sub>3</sub> *I <sub>2</sub>	284.10 ± 5.22	8.05 ± 1.31	8.66 ± 1.00	109.05 ± 3.76
S <sub>3</sub> *I <sub>3</sub>	279.70 ± 5.97	8.75 ± 1.77	8.39 ± 0.20	103.60 ± 13.83

Note: Averages with different letters in the same column have significant difference (P<0.05)

Table 2. Weight of leaf (g), stem (g), and panicle (g)

Treatment	Sorghum		
	Leaf weight	Stem weight	Panicle weight
Sorghum			
S <sub>1</sub>	77.64 ± 17.61 <sup>a</sup>	290.10 ± 37.94 <sup>a</sup>	43.80 ± 7.79 <sup>a</sup>
S <sub>2</sub>	85.09 ± 19.62 <sup>a</sup>	317.00 ± 34.43 <sup>a</sup>	49.23 ± 11.06 <sup>a</sup>
S <sub>3</sub>	110.89 ± 26.18 <sup>b</sup>	447.03 ± 88.34 <sup>b</sup>	74.49 ± 15.39 <sup>b</sup>
Indigofera			
I <sub>0</sub>	98.25 ± 31.05	389.33 ± 116.36 <sup>a</sup>	61.83 ± 22.37
I <sub>1</sub>	87.47 ± 22.09	334.22 ± 61.46 <sup>b</sup>	54.92 ± 14.82
I <sub>2</sub>	89.75 ± 27.55	345.15 ± 99.50 <sup>ab</sup>	54.65 ± 17.89
I <sub>3</sub>	89.35 ± 21.95	336.80 ± 72.96 <sup>ab</sup>	51.95 ± 15.95
S vs I			
S <sub>1</sub> *I <sub>0</sub>	77.25 ± 19.08	300.59 ± 53.89	45.50 ± 8.49
S <sub>1</sub> *I <sub>1</sub>	82.65 ± 16.36	315.00 ± 32.97	44.40 ± 9.27
S <sub>1</sub> *I <sub>2</sub>	76.00 ± 22.96	276.25 ± 21.07	42.25 ± 9.37
S <sub>1</sub> *I <sub>3</sub>	74.65 ± 18.50	268.40 ± 30.05	43.05 ± 7.00
S <sub>2</sub> *I <sub>0</sub>	85.80 ± 19.44	334.15 ± 22.94	51.70 ± 17.65
S <sub>2</sub> *I <sub>1</sub>	81.80 ± 12.59	314.50 ± 4.16	50.60 ± 10.92
S <sub>2</sub> *I <sub>2</sub>	80.75 ± 22.12	303.80 ± 64.15	47.10 ± 8.24
S <sub>2</sub> *I <sub>3</sub>	92.00 ± 28.19	315.55 ± 25.14	47.50 ± 9.58
S <sub>3</sub> *I <sub>0</sub>	131.70 ± 22.51	533.10 ± 64.32	88.30 ± 4.92
S <sub>3</sub> *I <sub>1</sub>	97.95 ± 33.81	373.15 ± 98.57	69.75 ± 11.58
S <sub>3</sub> *I <sub>2</sub>	112.50 ± 26.76	455.40 ± 83.22	74.60 ± 14.36
S <sub>3</sub> *I <sub>3</sub>	101.40 ± 11.75	426.45 ± 20.62	65.30 ± 20.56

Note : Averages with different letters in the same column have significant difference (P<0.05)

Table 3. Biomass yield (ton/ha) of sorghum with indigofera in intercropping system

% Indigofera	Variety of sorghum			Average
	Patir 3.2	Patir 3.7	Citayam	
0	10.59 ± 1.33	11.07 ± 1.65	12.76 ± 4.64	11.47 ± 1.44 <sup>a</sup>
30	13.38 ± 3.49	14.25 ± 1.97	14.41 ± 3.00	14.01 ± 2.66 <sup>b</sup>
40	13.23 ± 2.09	14.77 ± 2.11	16.02 ± 1.99	14.67 ± 2.22 <sup>b</sup>
50	15.96 ± 1.32	17.15 ± 3.07	16.00 ± 3.17	16.44 ± 2.47 <sup>b</sup>
Average	13.29 ± 2.79	14.31 ± 3.02	14.80 ± 2.57	

Note : Averages with different letters in the same column or line have significant difference(P<0.05)

Two plants grown close together would not compete with each other, and the growth would not be disturbed if requirements for water, nutrients and light were met. However, at a certain point, a diminishing return point is reached. This is attained when plant density is too high, resources for growth are limited and light interception is reduced. There was large genetic variability among and within different races in the diversity panel for growth, physiological traits and yield components (Mutava and College, 2006)

Crop production is affected greatly by the environment in which they grow. The data showed that varieties of sorghum citayam produced the best height of plant than patir 3.2 and patir 3.7. This means that citayam variety was optimal. The highest growth plant was citayam variety because ability in performing cell. Biomass is an accumulation of various food reserves. More biomass a plant then the metabolic processes in plants running well and indicate the amount of nutrient that plants absorbed while biomass bit indicates a bottleneck in the process of metabolism.

Plant height showed significant differences among variety. Plant height is one indicator of growth as well as the parameters used to measure growth by environmental influences, since growth is the variables most easily seen and measurements can be carried out without damaging the plant samples. The process of plant height is preceded by the formation of buds which is a process of cell division and enlargement, influenced by cell turgor. The process of cell division and enlargement will happen when the cells undergo turgiditas the main element is the availability of water. Reduced supply of water causes turgiditas plant cells decreased even disappear. Turgiditas loss will inhibit cell division and enlargement.

The leaves have reached the maximum number of ranges 6-12 strands so that nutrients provided is intended for seed formation. Upon entering the generative growth of food reserves will be directed to the formation of the result of the plant. Availability of sufficient nutrients during growth causes the photosynthesis process will be more active, so that elongation, cell division and differentiation will be better accelerate leaf blade length. Photosynthesis active radiation absorbed by the intercropping was higher than sole crop (Rajali and DahMardeh, 2014)

Differences in growth characters may be due to the differences in genetic structure, in addition to the widely differences between genotypes for mineral elements concentrations and to the cultivar differences in partitioning of photosynthesis among plant organ. On the other hand, the inconstant line sorghum cultivar in leaf weight in their variation with advancing plant age could be attributed to the cultivar differences in migration of dry matter from vegetative organ to panicle and also to cultivar differences in photosynthesis partitioning (Hassanein *et al.*, 2010).

The differences of productivity components might be attributes to the variation in translocation rate of photosynthesis from leaves to the storing organs i.e. grains. It is noteworthy to mention that the results of cultivars differences in yield and its components may be due to the differences in genetic structure between sorghum cultivars and also the cultivars differences in photosynthesis partitioning. Again the widely differences between genotypes for mineral concentrations can be also participate in the cultivars differences in yield and its components (Hassanein *et al.*, 2010). There was large genetic variability among and within different races in the diversity panel for growth, physiological traits and yield components (Mutava and College, 2006).

## Conclusion

Varietas citayam have highest criteria for growth and productivity. The suitable variety of sorghum and indigofera for biomass yield was found in plots cultivated combination of patir 3.2 with 50 % indigofera, patir 3.7 with 50 % indigofera, and citayam with 40 % indigofera. The highest biomass yield was found in plots cultivated combination patir 3.7 with 50 % indigofera.

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# Herbage Production and Nutritive Value of Some Forage Legumes as Calf Feed Supplement

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## Abstract

Four forage legumes were evaluated in this experiment for their herbage production and nutritive values as calf feed supplement. The legumes i.e. *C. pubescens*, *C. ternatea*, *C. pascuorum* and *V. umbellata* were randomly planted in six replicates of 2x2 m<sup>2</sup> plot following a completely randomized design. Sowing was done by broadcasting seeds into the respective plot at 20 x 20 cm<sup>2</sup> and covered with soil by light hand raking. Variables including plant growth and biomass production, nutrient content and in vitro organic matter digestibility (IVOMD) were measured. *C. ternatea* was significantly ( $P < 0.001$ ) growing faster and producing higher amount of herbage compared to the other legumes. Herbage leaf: stem ratio was the highest ( $P < 0.01$ ) in *V. umbellata* and the lowest in *C. pascuorum*. Crude protein and crude fiber content of those forage legumes varied narrowly between 17 to 23% and 11 to 18% respectively. All four legumes had relatively high IVOMD varying between 78-83%. *C. pubescens* had significantly lower ( $P < 0.001$ ) IVOMD compared the other legumes. Total volatile fatty acid (VFA) was the higher ( $P < 0.05$ ) in *C. pascuorum* compared to the other legumes but proportion of acetate (C2), propionate (C3), butyrate (C4) and the ratio C3: (C2+C4) were comparable ( $P > 0.05$ ) among legumes. It can be concluded that *C. ternatea* was the best candidate as the base of calf supplement.

**Keywords:** biomass production, calf supplement, forage legumes, nutritive value

## Introduction

Despite considerable attempts have been put by the Indonesian government to boost beef production, cattle productivity remains low as a result the substantially high calf mortality rate and slow growth rate during pre-weaning period. Low growth rate during this period has been proven to impact on low weaning rate, growth rate during the later stage of growth and thereafter selling weight. Supplementation of calves during the dry season before weaning has proved to be a promising option to substantially improve beef cattle production in the Province of East Nusa Tenggara, with potential benefits for famers. Provision of a small amount (2% liveweight, LW) of locally blended concentrate supplement to Bali calves before weaning can significantly reduce mortality rate (from 20 to 50% to less than 1%) and increase growth rate (206 versus 100 g/day live weight gain) (Jelantik *et al.*, 2008). The yearling weight for supplemented calves was almost double that of the control calves (Copland *et al.*, 2011).

Farmer adoption for this technology was however considerably low (Parker *et al.*, 2012). The unavailability of feed supplement on market, reasonably high price, processing requirement and application that require temporary calf separation from their dams were thought among the reason for the low adoption rate. Therefore, there is a need to improve adoption rate of calf supplementation through incorporating herbaceous legumes in the formulation of calf supplement thereby reducing the supplement price and can be produced by farmer. The experiment was conducted to screen some promising herbaceous legumes as calf supplement.

## Materials and Methods

Four forage legumes including *C. pubescens*, *C. ternatea*, *C. pascuorum* and *V. umbellata* were randomly planted in six replicates of 2x2 m<sup>2</sup> plot following a completely randomized design. Sowing was done by broad casting seeds into the respective plot at 20 x 20 cm<sup>2</sup> and covered with soil by light hand raking on 9 July 2014. The trial was carried out under field conditions at Noelbaki, (10° 05'S and 123° 52' E at an altitude of 10 asl), Kupang regency, East Nusa Tenggara Province, Indonesia. The soil type at the site is a vertisol (Typic Ustropepts/Septicols) with deep soils (50-60 cm) and a pH of 7.7.

The growth of four legumes species was measured be-weekly by measuring the height from the ground to the highest point of the plant for the duration of the experiment. The biomass production was determined by harvesting forage from sample plots at about three months after planting at vegetative or at the strat of generative stage. Following harvest, the biomass of each species was freshly weighed and sub-samples were oven-dried at 105 °C to a constant weight between 3 to 6 hours to record the dry matter production. Herbage samples were further determined for their nutritional content including crude protein, crude fiber, crude fat, and nitrogen free extract following the proximate analyses. The procedure proposed by Tilley and Terry (1963) was followed to estimate *in vitro* dry matter (IVDMD) and organic matter digestibility (IVOMD).

The experimental data were subjected to analysis based on a Randomized Complete Design using a GLM procedure according to the SPSS18 program. Means were compared using LSDs.

## Results and Discussion

Results showed that *C. ternatea* was among the fastest growing forage legumes producing the highest herbage (Table 1). This typically fast growing pattern of *C. ternatea* is required for successful biomass production in area like EAT where rainy season last in a short period of 3-4 months. It also promised reasonable forage production when planted after corn harvest to utilize the remaining soil moisture during the end of rainy season. With biomass production reaching 6 ton DM/ha when harvested nearly flowering stage, *C. Ternatea* could be utilized as for dry season supplement for staggering 66 calves when offered at 2% BW (Copland *et al.*, 2011). Moreover, some agronomic characteristics of the legumes other legumes under investigation in this experiment indicated that herbage produced was of reasonably good quality, e.g. by the very high leaf:stem ratio. Leaf contain much higher crude protein compared to stem (Van Soest, 1994). In addition, leaves contain less fibre and those fibres are more degradable than that in stem (Akin, 1984). Result of chemical analyses confirmed that all legumes contain reasonably high crude protein varying from 15.87% in *C. ternatea* to 23.42% in *C. vascurum*. All legumes contain less than 20% crude fibre.

Table 1. Agronomic characteristics and biomass production of forage legumes harvested at vegetative stage

Variables	Herbaceous Legumes				MSE	P-value
	<i>C. pascuorum</i>	<i>C. pubescens</i>	<i>C. ternatea</i>	<i>V. umbellata</i>		
Height (cm)	12.33ab	8.75a	44.88c	16.50b	9.05	<0.001
Branch number	9.33c	2.50b	8.75c	0.00a	1.49	<0.001
Leaf no.	35.33b	9.50a	51.50c	5.50a	27.97	<0.001
Leaf proportion (% DM)	65.97a	77.23bc	71.52ab	84.79c	0.004	0.008
Stem proportion (%DM)	34.03c	22.77ab	28.47bc	15.21a	0.004	0.008
Herbage production (ton/ha)	1.03a	0.62a	6.05b	1.69a	8.365	<0.001

Values followed by different superscripts within the same row showed significant different (P<0.05) MSE=mean square error, P=probability

Rumen digestibility *in vitro* is a reasonably good indicator for the availability of forages for rumen fermentation. Results showed that almost all legumes had high rumen digestibility varying from 62% up to 80%. This rumen digestibility is higher than that of grasses, tree legumes and bushes (Jelantik, 2001). This could be an indication for a more suitable use of those legumes as calf supplement. With rumen that still under development, calf supplement should be highly digestible and has high biological value (Davis and Drackely, 1998; Donnelly dan Hutton, 1976), high ME utilization, i.e. 86% (Gerrits *et al.*, 1996), without heat treatment (Wilson dan Wheelock, 1972) and containing no anti-nutritive factor (ANF) (Lalles, 1993). Herbaceous legumes are known to have negligible ANF (Van Soest, 1994). Results also showed that IVOMD as an estimate for the energetic value (Van Soest, 1994) of all legumes were relatively high, i.e. over 78% for all legumes under study. The highest IVOMD (P<0.001) were in *C. ternatea* and *V. Umbellata* which reached a level more than 83%. This indicated high potency to be utilised as the base of calf supplement.

Table 2 showed the concentrations of fermentative products in the liquid residues taken during the determination of *in vitro* dry matter digestibility of four forage legumes. Volatile fatty acids (VFA) concentration in the residues were relatively high in all legumes but total VFA concentration was higher

( $P < 0.05$ ) in *C. vascuorum* compare to other legumes. Higher concentration of VFA would stimulate better rumen development (Davis and Drackely, 1998). This could indicate that *C. vascuorum* would stimulate better rumen development of Bali calves when used as a supplement. However, the proportion of acetate to propionate and butyrate is also important in the stimulation of rumen development as both propionate and particularly butyrate are better stimulant than acetate. In this case all legumes produced comparable proportion individual acids as well as their ratio which indicated all legumes would comparably stimulate rumen development.

Table 2. Means of volatile fatty acids (VFA) concentration in residues of in vitro dry matter digestibility

Rumen Variables	<i>C. vascuorum</i>	<i>C. Ternatea</i>	<i>V. Umbellata</i>	<i>C. pubescens</i>	MSE	P-value
Concentration (mmol/L):						
Acetate	84.61a	52.55b	55.53b	58.49b	136.52	0.046
Propionate	27.34	17.45	16.09	17.38	39.42	0.244
Butyrate	10.65	7.66	8.15	7.07	4.122	0.170
Total VFA	122.61b	77.65a	79.77a	82.95a	317.98	0.067
Proportion (%):						
Acetate	69.37	67.43	69.60	70.90	14.539	0.219
Propionate	22.07	22.30	20.07	20.40	13.913	0.611
Butyrate	8.57	10.27	10.33	8.70	4.051	0.284
Rasio propionate : (acetate+butyrate)	0.285	0.287	0.253	0.260	0.003	0.585

Values followed by different superscripts within the same row showed significant different ( $P < 0.05$ ). MSE=mean square error, P=probability

## Conclusion

All legumes contained nutrient suitable for calf supplement. However, the fast growing and high biomass production of *C. ternatea* offers the best candidate for formulation of forage based calf supplement. This promises high adoption rate by small scale farmer.

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# Evaluation of Growth and Biomass Production of Sorghum Mutant Lines (Sorghum Brown midrib) at Different of Harvest Time

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## Abstract

*The aim of this experiment was to evaluate the growth and production of sorghum brown midrib (BMR) lines at different of harvest time. This research was conducted from June to October 2014 at SEAMEO Biotrop Bogor. manure and NPK were used as fertilizer. The design of this experiment was completely block design with three replications. The first factor was the BMR sorghum lines (Patir 3.1 (as control), Patir 3.2, Patir 3.3, Patir 3.4, Patir 3.5, Patir 3.6 and Patir 3.7), the second factor was the harvesting time (flowering stage, soft dough stage and hard dough stage). Parameters measured were plant height, leaf width, leaf length, stem, percentage of leaves and stems and sugar content. If significantly different effect founded continue to further test Duncant. The results showed that the harvesting time had a significant effect on plant height, whereas the BMR lines and harvest time significantly different effect on leaf width. There is an interaction between the BMR lines and the harvesting time on stem diameter, leaf length, the percentage of stems and leaves and sugar content. While this BMR lines and time of harvest significant effect on the fresh and dry matter production. Strain Patir 3.2 produces the highest dry matter production.*

**Keywords:** growth, production, sorghum brown midrib lines

## Introduction

Sorghum is a crop that has potency to become material of sorghum forage (green plants) because high nutrient content, palatable and high biomass production. Sorghum plant breeding with the application of nuclear technology through gamma ray radiation has produced some Sorghum Brown Midrib lines. Sorghum Brown midrib (Bmr) has a lower lignin content ( $\pm 4\%$ ) than conventional sorghum, so expect the Bmr sorghum would have a better digestibility. Lignin content and composition of the cell wall of forage is affected by plant maturity. The level of plant maturity will affect forage production and nutritive value. Determination of harvesting time for Bmr sorghum lines is necessary to know the best line and harvesting time on growth and biomass production of Bmr sorghum.

## Materials and Methods

This research was conducted in area of  $\pm 1500\text{ m}^2$  at SEAMEO Biotrop research garden Bogor. Fertilizer were used urea, TSP, KCl with a ratio of 4:3:2 (g/g/g) with a total of 270 kg/ha. The second fertilizer at the age of 50 DAS (days after seeding) were used Urea, TSP, KCl with a ratio of 2:4:2 (g/g/g) with a total of 200 kg/ha. Pest birds prevention was used plastic bags 1 kg size that has been hollowed out.

This study used a randomized block design factorial pattern with three replications. The first factor were the Bmr sorghum lines (Patir 3.1 (as a control), Patir 3.2, Patir 3.3, Patir 3.4, Patir 3.5, Patir 3.6 and Patir 3.7), while the second factor was the time of harvest (flowering, soft dough and hard dough phase). If there was a significant effect then conducted a further test used Duncant test.

## Results and Discussion

### Fresh and Dry Material Production

The research data showed that there was no interaction between the time of harvest with Bmr sorghum lines on fresh and dry matter production. But lines and harvesting times affected of fresh and dry matter production of sorghum. From Table 1 it could be seen that fresh and dry matter production lines P 3.7 and P 3.2 were not different compared to control (P 3.1). This suggests that the ability of crop production Bmr

sorghum (P 3.2 and P 3.7) were equal to the non Bmr sorghum production (P 3.1). The same result was reported by Kurniawan (2014) that the Bmr sorghum lines (P 3.2 and P 3.5) produced of dry matter not different from sweet sorghum varieties Citayam and Numbu.

Table 1. Mean of fresh and dry matter production of sorghum (g)

Lines	Parameter	Harvesting Times			Mean
		Flowering	Soft Dough	Hard Dough	
P 3.1	Fesh Prod	3782.6±213.49	4108.61±560.90	4556.6±195.28	4149.27±323.22A
	DM Prod	2549.08±161.58	2542.57±339.75	2187.82±105.95	2426.49±202.43a
P 3.2	Fesh Prod	3996.9±197.80	4075.47±612.38	3970.26±381.72	4014.21±397.30A
	DM Prod	2715.66±181.45	2434.50±407.19	2070.61±279.73	2406.92±289.46a
P 3.3	Fesh Prod	3193.79±75.41	3456±626.57	3745±158.82	3464.93±286.93B
	DM Prod	2166.48±118.89	1959.59±369.11	1876.48±104.70	2000.85±197.57b
P 3.4	Fesh Prod	2909.1±144.04	3767.53±219.26	3703±506.71	3459.87±290.01B
	DM Prod	1849.65±105.31	2185.74±106.35	1895.02±279.05	1976.80±163.57b
P 3.5	Fesh Prod	3191.3±175.67	3686.75±625.10	3590.66±303.74	3489.57±368.17B
	DM Prod	2090.01±109.42	2102.93±357.23	1709.87±68.28	1967.60±178.31b
P 3.6	Fesh Prod	3404.06±301.73	3542.20±277.91	3911±267.95	3619.09±282.53B
	DM Prod	2271.01±233.78	2015.70±185.34	2003.36±93.81	2096.69±170.98b
P 3.7	Fesh Prod	3544.26±376.94	4387.15±422.83	4392.33±499.50	4107.91±433.09A
	DM Prod	2336.89±387.31	2750.18±148.36	2335.15±208.87	2474.07±248.18a
Mean	Fesh Prod	3431.71±212.16B	3860.53±477.85A	3981.26±330.53A	
	DM Prod	2282.68±185.39a	2284.46±273.33a	2011.19±162.91b	

Description: Superscript (uppercase) in the same row and column shows the significant. Superscript (lowercase) in the same row and column shows the significant.

In general, the harvest time gave significantly effect on fresh and dry matter production. Hard dough and soft dough stage were give higher dry matter production than the flowering stage. These were because the soft dough and hard dough stage on filling stage so that the seeds were increase grain weight and generally increased the weight of the plant. According to Vanderlip (1993) the emergence of seed would increase the weight of the plant, then pollination, seed weight would increase and followed by dry matter production.

### Ratio Leaves and Stem

Results showed that there were an interaction between the lines and the time of harvest on the ratio leaf and stem. It could be seen that the ratio of leaf lines P 3.1, P 3.2, P 3.3, P 3.4, P 3.5, P 3.6 and P 3.7 were not significant compared to the control (P 3.1). Table 2 it showed that in general the ratio of leaves would increase in the soft dough stage and decreased after the hard dough stage, as well as the percentage of stems. A decrease in the ratio of leaves and stems was due to the movement of nutrients from the leaves and stems to the seed. Gerik, et al., (1994) stated that after flowering, plant growth was concentrated in the seed formation. Sugars, amino acids and protein in the leaves, stems and roots would be transported quickly to the seed will then converted into starch and protein.

### Plant Height and Stem Diameter

Table 3 showed that there was no interaction between the lines and the time of harvest on plant height, and the harvest time significant effect on plant height. In order side, stem diameter has an interaction between the lines and the time of harvest, which contained the highest stem diameter at treatment lines P 3.7 at harvest time of flowering.

In generally, the harvest time significant increased plant height at soft dough stage and then decreased after hard dough stage. In contrast to the results of Qu et al (2014), that high plant of Hunningreen sorghum increased in maturity from stage milk stage to dough stage. These due to the sampled plants were different individuals so that there was variation in plant height parameters.

Table 2. Ratio of leaves, stem and panicle of sorghum (%)

Lines	Parameter	Harvesting Times			Mean
		Flowering	Soft Dough	Hard Dough	
P 3.1	% Leaves	29,20±1,49 <b>A</b>	29,36 ± 3,01 <b>A</b>	24,21 ± 0,25 <b>CD</b>	27,59 ± 1,58
	% Stem	19,37 ± 0,25 <b>d</b>	25,55 ± 2,89 <b>a</b>	15,00 ± 0,75 <b>ef</b>	19,97 ± 1,30
	% Panicle	51.44±1.63 <b>CDE</b>	45.09±0.53 <b>G</b>	60.79±0.74 <b>A</b>	52.44±0.96
P 3.2	% Leaves	25,43 ± 3.08 <b>CD</b>	28,58 ± 0,74 <b>A</b>	25,05 ± 0,37 <b>CD</b>	26,36 ± 1,40
	% Stem	19,34 ± 1,76 <b>d</b>	24,48 ± 0,62 <b>ab</b>	15,25 ± 2,30 <b>ef</b>	19,69 ± 1,56
	% Panicle	52.81±3.02 <b>BC</b>	46.93±0.47 <b>FG</b>	59.70±1.99 <b>A</b>	53.15±1.83
P 3.3	% Leaves	26,02 ± 5,24 <b>A</b>	27,44 ± 0,24 <b>ABCD</b>	25,43 ± 0,42 <b>CD</b>	26,29 ± 1,97
	% Stem	16,63 ± 0,76 <b>e</b>	23,48 ± 0,44 <b>abc</b>	15,51 ± 0,71 <b>ef</b>	18,54 ± 0,64
	% Panicle	54.37±0.44 <b>B</b>	49.09±0.26 <b>EF</b>	59.07±0.34 <b>A</b>	54.17±0.35
P 3.4	% Leaves	28,98 ± 0,67 <b>A</b>	25,68 ± 1,29 <b>CDE</b>	24,30 ± 1,25 <b>CD</b>	26,32 ± 1,07
	% Stem	20,09 ± 0,83 <b>d</b>	23,87 ± 0,44 <b>ab</b>	14,53 ± 0,60 <b>ef</b>	19,50 ± 0,62
	% Panicle	49.35±2.02 <b>DE</b>	50.44±1.67 <b>CDE</b>	61.17±0.78 <b>A</b>	53.65±1.49
P 3.5	% Leaves	28,98 ± 1,73 <b>A</b>	27,92 ± 0,06 <b>ABC</b>	25,04 ± 0,95 <b>CD</b>	27,32 ± 0,91
	% Stem	19,67 ± 1,08 <b>d</b>	22,92 ± 0,37 <b>bc</b>	15,72 ± 1,69 <b>ef</b>	19,43 ± 1,05
	% Panicle	51.35±0.72 <b>CDE</b>	49.16±0.32 <b>DEF</b>	59.24±1.03 <b>A</b>	53.25±0.69
P 3.6	% Leaves	27,20 ± 0,24 <b>ABCD</b>	29,50 ± 1,00 <b>A</b>	25,78 ± 1,08 <b>BCDE</b>	27,49 ± 0,77
	% Stem	20,51 ± 0,22 <b>d</b>	21,46 ± 0,82 <b>cd</b>	13,87 ± 0,18 <b>f</b>	18,61 ± 0,41
	% Panicle	52.29±0.42 <b>BC</b>	49.05±0.73 <b>EF</b>	60.35±0.91 <b>A</b>	53.90±0.69
P 3.7	% Leaves	28,05 ± 0,51 <b>ABC</b>	28,20 ± 0,88 <b>AB</b>	25,24 ± 1,40 <b>CD</b>	27,16 ± 0,60
	% Stem	20,42 ± 0,81 <b>d</b>	23,38 ± 1,84 <b>abc</b>	14,48 ± 0,86 <b>ef</b>	19,43 ± 1,17
	% Panicle	51.53±0.83 <b>CD</b>	49.47±1.22 <b>DE</b>	60.28±0.62 <b>A</b>	53.76±0.89
Mean	% Leaves	27,69 ± 1,85	28,10 ± 1,03	25,01 ± 0,67	
	% Stem	19,43 ± 0,82	23,59 ± 1,06	14,91 ± 1,01	
	% Panicle	51.88±1.30	48.46±0.74	60.08±0.92	

Description: Superscript (uppercase) in the same row and column shows the interaction. Superscript (lowercase) in the same row and column shows the interaction.

Table 3. Mean of Plant height (cm) and stem diameter (mm) of Sorghum

Lines	Parameter	Harvest times			Mean
		Flowering	Soft Dough	Hard dough	
P 3.1	Height	205,49 ± 2,81	219,46 ± 4,73	213,90 ± 0,51	212,95 ± 2,68
	Diameter	13,7 ± 1,32 <b>gh</b>	14,72 ± 1,02 <b>cdefg</b>	15,6 ± 0,3 <b>bcde</b>	14,53 ± 0,88
P 3.2	Height	207,04 ± 3,10	215,71 ± 6,13	212,08 ± 9,56	211,61 ± 6,26
	Diameter	17,41 ± 0,70 <b>ab</b>	14,32 ± 1,76 <b>defg</b>	15,43 ± 1,32 <b>cdef</b>	15,72 ± 1,26
P 3.3	Height	218,87 ± 5,50	216,89 ± 0,89	214,46 ± 8,13	216,74 ± 4,84
	Diameter	12,16 ± 1,68 <b>h</b>	13,85 ± 0,78 <b>defg</b>	14,97 ± 0,29 <b>cdefg</b>	13,66 ± 0,92
P 3.4	Height	213,62 ± 0,27	220,75 ± 3,08	213,37 ± 0,92	215,91 ± 1,42
	Diameter	12,4 ± 2,40 <b>h</b>	13,03 ± 0,29 <b>gh</b>	15,37 ± 1,41 <b>cdef</b>	13,6 ± 1,37
P 3.5	Height	217,05 ± 4,62	217,18 ± 2,57	215,09 ± 5,74	216,44 ± 4,31
	Diameter	14,83 ± 0,32 <b>cdefg</b>	13,5 ± 1,39 <b>fgh</b>	14,9 ± 0,26 <b>cdefg</b>	14,41 ± 0,66
P 3.6	Height	220,20 ± 3,50	217,83 ± 3,43	215,28 ± 1,43	217,77 ± 2,79
	Diameter	15,63 ± 0,21 <b>bcde</b>	14,6 ± 0,75 <b>cdefg</b>	16,4 ± 0,72 <b>abc</b>	15,54 ± 0,56
P 3.7	Height	214,18 ± 4,21	214,20 ± 4,04	214,76 ± 8,83	214,38 ± 5,69
	Diameter	17,35 ± 0,49 <b>a</b>	15,93 ± 0,41 <b>bcd</b>	16,2 ± 1,3 <b>abcd</b>	16,49 ± 0,74
Mean	Height	213,78 ± 3,43 <b>B</b>	217,43 ± 3,55 <b>A</b>	214,13 ± 5,12 <b>B</b>	
	Diameter	14,72 ± 1,01	14,28 ± 0,91	15,55 ± 0,80	

Description: Superscript (uppercase) in the same row and column shows the significant. Superscript (lowercase) in the same row and column shows the interaction.

## Width and Length Leaves

Table 4. Mean of width and length leaves of sorghum (cm)

Sorghum Lines	Parameter	Harvest Times			Mean
		Flowering	Soft Dough	Hard dough	
P 3.1	Width	7,61 ± 0,27	7,43 ± 0,50	7,88 ± 0,40	7,64 ± 0,39 <b>AB</b>
	Length	99,83 ± 7,73 <b>abc</b>	100,98 ± 2,69 <b>abc</b>	103,85 ± 1,13 <b>a</b>	101,55 ± 3,85
P 3.2	Width	7,50 ± 0,26	6,89 ± 0,45	7,56 ± 0,39	7,32 ± 0,37 <b>BC</b>
	Length	99,81 ± 1,08 <b>abc</b>	97,74 ± 2,42 <b>bcd</b>	96,56 ± 2,78 <b>bcd</b>	98,04 ± 2,09
P 3.3	Width	6,99 ± 0,20	6,84 ± 0,72	7,93 ± 0,21	7,25 ± 0,39 <b>C</b>
	Length	97,58 ± 1,91 <b>bcd</b>	97,61 ± 1,98 <b>bcd</b>	93,45 ± 1,99 <b>de</b>	96,21 ± 1,96
P 3.4	Width	6,64 ± 0,33	6,82 ± 0,24	7,68 ± 0,63	7,04 ± 0,40 <b>C</b>
	Length	96,85 ± 4,07 <b>bcd</b>	97,36 ± 2,64 <b>bcd</b>	95,6 ± 3,22 <b>cd</b>	96,60 ± 3,31
P 3.5	Width	7,16 ± 0,01	6,77 ± 0,51	7,36 ± 0,24	7,10 ± 0,25 <b>C</b>
	Length	98,83 ± 0,55 <b>abcd</b>	97,94 ± 1,71 <b>bcd</b>	96,47 ± 2,25 <b>bcd</b>	97,74 ± 1,50
P 3.6	Width	7,35 ± 0,21	6,76 ± 0,21	7,67 ± 0,29	7,26 ± 0,24 <b>C</b>
	Length	100,00 ± 1,70 <b>abc</b>	98,76 ± 0,58 <b>abcd</b>	90,67 ± 4,78 <b>e</b>	96,48 ± 2,35
P 3.7	Width	7,56 ± 0,20	7,68 ± 0,36	8,04 ± 0,54	7,76 ± 0,37 <b>A</b>
	Length	101,49 ± 1,94 <b>ab</b>	100,62 ± 0,70 <b>abc</b>	101,72 ± 3,24 <b>ab</b>	101,28 ± 1,96
Mean	Width	7,26 ± 0,21 <b>B</b>	7,03 ± 0,43 <b>C</b>	7,73 ± 0,39 <b>A</b>	
	Length	99,20 ± 2,71	98,72 ± 1,81	96,90 ± 2,77	

Description: Superscript (uppercase) in the same row and column shows the significant. Superscript (lowercase) in the same row and column shows the interaction.

The data showed there was no interaction between the lines and harvest time on leaf width. But the lines and time of harvest influence sorghum plant leaf width. The widest leaves produced from line P 3.7 while harvest time produces the widest leaves are hard dough stage. The results showed that leaf width line P 3.7 was not significantly different from controls.

Table 4 showed an interaction between the lines and the time of harvest of the length leaves sorghum. Strain P 3.7 at the hard dough stage produces leaf length was not significantly different than the control (P 3.1).

## Sugar Stems

The research results showed an interaction between the lines and the time of harvest on sorghum stem of sugar brix levels. The highest levels of brix sugar was contained in P 3.3 at hard dough stage.

Table 5. Mean of sugar stems of sorghum (% brix)

Lines	Harvest Times			Mean
	Flowering	Soft Dough	Hard dough	
P 3.1	10,67 ± 0,72 <b>G</b>	12,37 ± 0,49 <b>DEF</b>	11,63 ± 1,38 <b>EFG</b>	11,56 ± 0,86
P 3.2	11,28 ± 0,59 <b>FG</b>	14,77 ± 0,42 <b>BC</b>	15,8 ± 0,61 <b>AB</b>	13,95 ± 0,54
P 3.3	12,85 ± 0,35 <b>DE</b>	14,8 ± 0,4 <b>BC</b>	16,7 ± 0,92 <b>A</b>	14,78 ± 0,55
P 3.4	12,28 ± 0,44 <b>DEF</b>	14,77 ± 0,57 <b>BC</b>	14,83 ± 0,32 <b>BC</b>	13,96 ± 0,44
P 3.5	12,47 ± 0,38 <b>DEF</b>	13,5 ± 0,36 <b>CD</b>	15,03 ± 1,59 <b>B</b>	13,67 ± 0,78
P 3.6	12,27 ± 0,25 <b>DEF</b>	13 ± 0,3 <b>DE</b>	14,43 ± 0,81 <b>BC</b>	13,23 ± 0,45
P 3.7	12,27 ± 0,17 <b>DEF</b>	12,6 ± 0,1 <b>DEF</b>	15,23 ± 1,44 <b>B</b>	13,37 ± 0,90
Mean	12,01 ± 0,56	13,68 ± 0,38	14,81 ± 1,00	

Description: Superscript (uppercase) in the same row and column shows the interaction.

In general, sugar brix levels increased with advanced maturation. Sugar content in sorghum stem was affected by maturity. The maturity of the stem was higher so the sugar content is also higher. Maturity sorghum stem with a maximum sugar content reached physiological maturity phase. This was consistent with the statement by Long et al (2006) that the sugar content in two varieties of sweet sorghum low at the beginning of flowering then increased gradually from early milky stage to seed maturation phase.

## Conclusion

Lines P 3.2 and P 3.7 produce the highest dry matter production at any time of harvest as same as the control (P 3.1).

## Acknowledgement

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# Dynamic Respons of Forage Availability to Landuse Exchange in Bogor Regency

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## Abstract

*One of important component in ruminant livestock is forage availability to supply feed sufficient. We reviewed recent data of importance to understanding forage availability response to land use exchange in Bogor Regency. Topics included plant productivity response to various land type, interactions with land type variables and ecological condition, impacts of increased landuse exchange and frequency of extreme conditions, animal number and trading flow, issues in biodiversity and vulnerability of land for pasture. We critically analyze the links between fundamental knowledge at farmer level and the additional land-economics variable that determine actual forage production and trade of feed at local scales. We found significant differences in plant biomass and plant nitrogen content between 2009-2011. We concluded that forage availability in Bogor will be decrease a long landuse exchange due to most of agriculture land changes to resident or infrastructure area. We recomended that forage needs could be fulfilled by focus on continued and improved integration of forage and horticulture cultivation.*

*Keywords: availability, Bogor, forage, landuse, modelling*

## Introduction

Bogor is one of important buffer region in supplying daily livestock product for capital city Jakarta Ruminant population in Bogor including beef cattles, cows, buffalos, goats and sheepsreached 385 571 heads (BPS, 2013), f . In order to sustain the supply of animal products, productivity of kept animals in Bogor has to be supported by feed availability, which in turn is associated with land use system. In this case, exchangable land use may have important role in provision of feed materials, in particular forages. Forage could be supplied by grass, legume or horticultural waste, such as plant parts or weeds. Forage yield maps coupled with site-specific in application of soil amendmets, fertilizers, and pesticides will allow the stakeholders to determine the forage availability for livestock management decisions (Digman and Shinnars, 2012). Land management for food and feed production is a fundamental human activity to support the livelihood. Understanding the condition and changes through time of the globally valuable land resource is important for forage supply in Bogor. For example, land-cover and land-use change can potentially affect the number of natural grassland or agriculture area.

Land use implies a human dimension or purpose for which the land is used (Lambin et al., 2001). Accurate information on land use is critical for understanding the causes of land cover change and for developing effective policies and strategies to slow and reverse agriculture land loss. The aim of this research was to understanding forage availability response to land use exchange in Bogor Regency.

## Materials and Methods

Data profiles including land use, dynamic population of animals, geographical maps and social economics data in Bogor region were obtained from Agriculture and Forest Office (2009-2013); Livestock and Fisheries Office (2009-2014); Spatial and Land Office (2010); Plan Map of Spatial and Region (2005-2025); and Statistic Centre Agency (2009-2010).

To model future forage availability additional datasets were compiled, which represents land use changes from forage sources area such as natural pasture, paddy fields, swamps, community forests, plantations, dry farms, yards, unirrigated areas, fallow paddy fields, and dry area. They include location of villages (counted in the census), roads, location of protected forests and suitability of soils for agriculture. We also collected some ruminants datasets to model future forage needs, i.e.: number of ruminants, birth and mortality rate, slaughter rate, and ruminant's flow in Bogor.

We used descriptive data analyses (Mattjik and Sumartjaya 2000) to evaluate relation between forage sources area with forage consumption based on number of ruminant. The application of the model was carried out on a spatial aggregated level due to computation time (regression and modelling) and due to lack of detailed spatial drivers.

## Results and Discussion

### Landuse configuration in Bogor

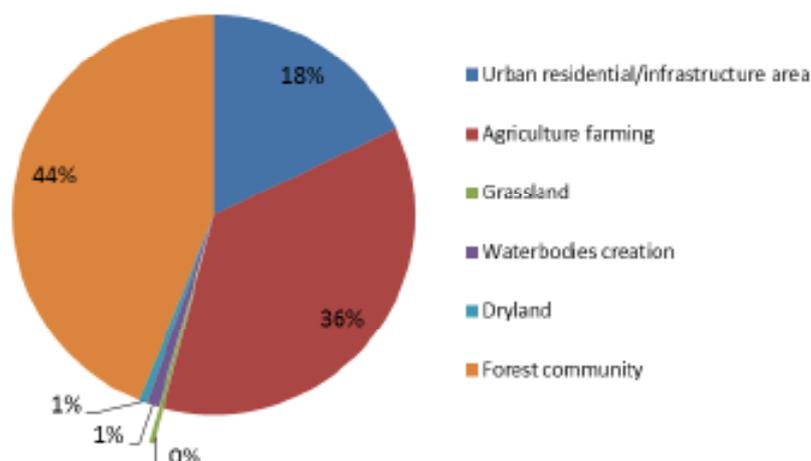


Figure 1. Landuse configuration in Bogor

The study area in Bogor district was characterized by a high fraction of forest community, agricultural areas and community regency (Fig. 1). The location of the huge areas is determined by Bogor city and Bogor Botanical Garden as protected regions, mainly for trees. Two big mountain areas, Salak and Pangrango, are in the very south of the catchment and conserved forest. In the northwestern part, Jonggol, extended areas with long lasting agricultural and grassland/natural pasture production are found. But there are also regions with scattered fields in remote areas with obviously land converted to community residents, unfortunately.

### Forage dynamics

In Bogor, most farmer uses forage from grass, legume and some trees for their animal, in small portion of agricultural crops is given for dairy cattle. Unfortunately there are no reliable data about the production of agricultural crops available to estimate intensification of production. Setiana (2014) stated that community uses natural plants as forage from resident areas, vacant lands, roadsides, riverbanks and others. In our analyses, we found the main forage sources in Bogor are meadow fields, garden, fallowed paddy field and vacant lands. There were a dynamic of forage production through landuse exchange and percentage of each forage sources.

Forage from grassland/natural pasture is not sufficient available for most farmers and intensification of grassland is on a very low level (Fig. 2). We critically analyze the links between fundamental knowledge at farmer level and the additional land-economics variable that determine actual forage production and trade of feed at local scales. We found significant differences in plant biomass and plant nitrogen content between 2009-2011. Through our field campaigns hardly any intensification is visible.

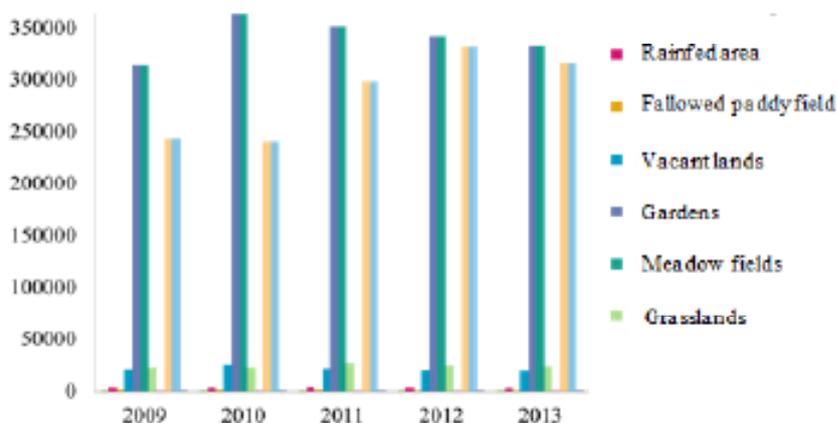


Figure 2. Dynamic of forage production from some sources (kg)

## Analysis of driving forces and conditions

The first step in setting up a model was the selection of driving forces that determine the configuration of land-use for forage and number of ruminant. As the main landuse change is the expansion of agricultural land modelling of future landuse is focused on this type of landuse. Our calculation resulted that Bogor has high potency to produce forage from some sources and supports by rice straw production.

The second step was connected forage availability and ruminant needs. Forage needs for ruminant based on population density with birth rate and mortality variable (Table 1). In other side, ruminant population in Bogor also has positive correlated with ruminant trade flow to support meat needs in Jakarta. It might caused a high forage needs for ruminant consumption and resulted the lack of forage to fulfil ruminant need.

Table 1. Forage production and ruminant needs based on datasets

Forage production and ruminant needs	Ton/Year				
	2009	2010	2011	2012	2013
Production of forage in sources area	621,335	671,938	757,160	774,634	743,430
Rice straw production	2.14	2.34	2.57	2.81	2.80
Forage needs	1,438,462	1,512,392	1,557,617	1,607,995	1,678,460
The lack of forage availability	-817,125	-840,451	-800,454	-833,358	-935,027

P: Production of forage in sources area (ton/year) = a x b x c

a: potencial area for forage production (ha)

b: forage production potency of each area (%)

c: natural forage production (ton/ha/year)

R: Rice straw production (ton/year) = rice yield production (ton/year) x 1.4 x forage utilization (%)

1.4: coefficient of rice straw yield from rice production system

F: Forage needs (ton/year) = ruminant population (head) x forage consumption (ton/head/year)

The lack of forage availability (ton/year) = (P + R) - F

## Conclusion

We concluded that forage availability in Bogor will be varied long landuse exchange and the number of ruminant was varied due to natural factors of animal population changes, ruminant trading flow, and the number of slaughtered ruminant.

We recommended that forage needs could be fulfilled by focus on continued and improved integration of forage and horticulture cultivation.

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**THEME D.**  
**ANIMAL NUTRITION**

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# Rain Tree Pod (*Samanea saman*) In Livestock Feeds: Opportunity, Challenges and Possibility

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## Abstract

The finding alternative natural feed sources could reduce livestock production costs. A rain tree pod (RTP) is a product from the rain tree, which is a tropical legume. It contained 766 g DM/kg as fresh-basis and it contained (g/kgDM), 87.3 g ash, 29.4 g nitrogen, 313 g neutral detergent fibre (NDF), 287 g acid detergent fibre (ADF), 52 g acid detergent lignin (ADL), 182 g total sugar, 84 g sucrose, 50 g phenolic compound, 35 g tannins, and 11 g condensed tannins. According to high total sugar and crude protein (CP) content, it is able to be used as a livestock feed. Therefore, incorporating rain tree pod in livestock diets will: (1) provide energy and protein sources, (2) provide phenolic compounds affecting to degradation of protein in the rumen (3) increase value of an agriculture product. Feeding trials aimed at evaluation the optimum amount of rain tree pod in feed, incorporation in microbial N production in the rumen, growth performance, milk production, and thus developing value-added in functional livestock feeds. The results obtained were: (1) feeding rain tree pods led to increasing microbial protein production in the rumen; (2) rain tree pods could be incorporated into the livestock feeds at 100 percent without affecting growth performance and milk quality; and (3) inclusion of rain tree pod led to significant reduction protein degradation in the rumen due to phenolic compounds component, it should be inactivated by adding poly ethylene glycol (PEG). Investigating factors that can not only enhance the feeding value, but also use for replacing molasses when requires for producing a bio-extract from some fruits/vegetable/herbs in order to save cost of using sugar and molasses and in the remote area which is hard to find molasses. This study provides a great potential for developing rain tree pods as a functional feeds and value-added feeds for livestock feed production/industry improvement.

**Keywords:** goats milk, growth rate, poly ethylene glycol (PEG), rain tree pods, rumen microbial N production

## Introduction

Rain tree pod is a product of the rain tree (*Samanea saman*). The plant can be usually grown on tropical climate area, with relatively low inputs and no irrigation. Although rain tree has been cultivated for multiple-purpose trees, such as curved wood, furniture, fertilizer, cultivation of lac (*Laccifer lacca Kerr*) and livestock fodder. Rain tree actually is not a forage plant; however, co-product (pod) obtained after ripening and falling down from tree is valuable as an animal feed (Jetana *et al.*, 2008; Staples and Elevitch. 2006.). To use rain tree pod as a potential animal feed requires information on its chemical composition, nutritive value, anti-nutritional agents, digestibility and product quality aspects. In the current report, opportunities, challenges and possibility with rain tree pod in the diet of goats are discussed.

## Chemical composition and nutritional value of rain tree pod

Rain tree pods collected from Nan Province, in the Northern region; Nakronpatham and Ayutthaya Provinces, in the Central region of Thailand for evaluating *in vivo* organic matter digestion (OMD) and estimating metabolizable energy (ME) contents by using gas production as described by Menke *et al.* (1979) procedures. The averaged values, it contained 766 g dry matter (DM)/kg as fresh-basis, 87.3 g ash, 29.4 g nitrogen, 313 g NDF, 287 g ADF, 52 g ADL, 182 g total sugar, 84 g sucrose, 50 g phenolic

compound, 35 g tannins, 11 g condensed tannin, respectively. Metabolizable energy for ruminants was 10 MJ/kg DM and OM digestibility was 872 g/kg DM.

### Feeding rain tree pod to animals

Consequently the results above, rain tree pod was used to evaluate for several studies with 3 reasons; (1) the high nutritive values with both protein and fermentable carbohydrate content, (2) containing high OMD and ME content, (3) rain tree is one of the multiple-purpose trees, being planted in Thailand; the rain tree pod is available in dry season and it is suitable to make a pellet feed and can be kept for a long time. Briefly, the pods of rain tree were firstly ground through a 10-14-mm grinding plate upon collection before subjected for second grinding using a 5-7-mm grinding plate equipped with 10-horsepower electrical meat grinder. The ground rain tree pods were sun dried for 12-18 h, and stored in air-tight container before used for analyses. The present report aims to evaluate and develop value-added in functional livestock feeds, (1) the rain tree pod pellets (RTPP) as a supplemental diet in *Saanen* dairy milking goats (2) the RTPP as a supplemental diet in *Black Bengal* goats, and (3) the RTPP adding with and without PEG as a supplemental diet in *Boer* crossed bred goats.

### Effects of feeding rain tree pods as a feed supplement to Saanen dairy goats on milk production and milk quality

The first experiment was conducted in 14 *Saanen* dairy goats in late lactation determined milk production and milk quality for 35 days. The animals were randomly divided into two groups of 7 animals each. Group 1 (38.4±0.62 kg) was fed *ad libitum* with 2.46 kg corn silage (fresh weight) and supplemented with 1 kg of commercial pellets (CMCP) (fresh weight), and group 2 (40.9±0.74 kg) was fed *ad libitum* with 2.46 kg corn silage (fresh weight) and supplemented with 1 kg of RTPP (fresh weight). Whilst the second experiment was used to determine the microbial N production in the rumen for 7 days (day 29-35). Table 1 shows that the average milk production (mL/day/BW<sup>0.75</sup>) and capital cost of milk production (US dollar/kg milk) were lower ( $P<0.05$ ), the contents of protein, lactose and total solids in the milk were greater ( $P<0.05$ ) in goats supplemented with RTPP than those supplemented with CMCP (Jetana *et al.*, 2012).

Table 1. Total OM intake and digestion, milk production, milk quality and microbial N production in the rumen in *Saanen* dairy goats late lactation fed corn silage and supplemented with commercial pellets (CMCP) and rain tree pod pellets (RTPP)

Dietary supplements	CMCP	RTPP	SED
Total OM intakes (g/BW <sup>0.75</sup> d <sup>-1</sup> )	79.2 <sup>b</sup> (n=7)	83.9 <sup>a</sup> (n=7)	0.02
Digestibility of OM	80.2 <sup>a</sup>	72.4 <sup>b</sup>	2.10
Milk Production (ml/d)	922 <sup>a</sup>	570 <sup>b</sup>	1.92
Milk produced capital (\$US/kg)	0.40 <sup>a</sup>	0.35 <sup>b</sup>	0.20
Chemical composition of milk (g/kg)			
Crude fat	45.6	42.1	4.62
Crude Protein	34.0 <sup>b</sup>	39.7 <sup>a</sup>	2.76
Lactose	23.3 <sup>b</sup>	34.7 <sup>a</sup>	2.94
Total ash	9.56	9.97	0.50
Solids non fat	64.0 <sup>b</sup>	84.4 <sup>a</sup>	2.69
Total solids	110 <sup>b</sup>	127 <sup>a</sup>	4.88
Total purine derivatives	27.4 <sup>a</sup>	17.6 <sup>b</sup>	1.92
PD (mmol)/kg DOMI	22.5 <sup>a</sup>	13.0 <sup>b</sup>	1.92
Microbial N (g N/day)	29.1 <sup>a</sup>	17.2 <sup>b</sup>	2.30

<sup>1</sup> Standard error of difference, <sup>2</sup> abcValues within the same column with different superscripts are significantly ( $P<0.05$ ) different. Values within the same column without different superscripts are not significantly ( $P<0.05$ ) different, *n*: numbers of animals, <sup>3</sup> using estimation model described by Jetana *et al.*, (2003)

### Effects of rain tree pods as a feed supplement to Black Bengal goats on ruminal microbial N production and growth performance

The first experiment was conducted in 8 *Black Bengal* goats determined the microbial N production in the rumen for 7 days (dietary adaptation 14 days). The animals were randomly divided into two groups

with 4 animals in each treatment group. Group 1 (22.5±0.72 kg) was fed *ad libitum* with chopped fresh *Napier* grass and supplemented with 0.5 kg of CMCP (fresh weight), and group 2 (23.1±0.84 kg) was fed *ad libitum* with chopped fresh *Napier* grass and supplemented with 0.5 kg of RTPP (fresh weight). Whilst the second experiment was used to determine growth rate and feed conversion rate for 72 days, the 12 male *Black Bengal* goats was randomly divided into two groups of 6 animals each. Group 1 (15.9±1.71 kg) was fed *ad libitum* with chopped fresh *Napier* grass and supplemented with 1 kg of CMCP (fresh weight), and group 2 (15.7±1.79 kg) was fed *ad libitum* with chopped fresh *Napier* grass and supplemented with 1 kg of RTPP (fresh weight). Table 2 shows that the average microbial N production in the rumen (mmol/day/BW<sup>0.75</sup>) and growth rate and feed conversion rate were lower ( $P<0.05$ ) in goats supplemented with RTPP than those supplemented with CMCP.

Table 2. Total OM intake and digestion, microbial N production and the growth performance in *Black Bengal* goats fed chopped fresh *Napier* grass and supplemented with commercial pellets (CMCP) and rain tree pod pellets (RTPP).

Dietary supplements	CMCP	RTPP	SED
1.1 Microbial N Production in the Rumen			
Total OM intakes (g/BW <sup>0.75</sup> d <sup>-1</sup> )	68.0 (n=4)	66.2 (n=4)	2.45
Digestibility of OM	72.2 <sup>a</sup>	62.9 <sup>b</sup>	2.09
Total purine derivatives	567 <sup>a</sup>	371 <sup>b</sup>	3.30
PD (mmol)/kg DOMI	11.6	10.1	1.58
Microbial N (g N/day)	6.52 <sup>a</sup>	4.97 <sup>b</sup>	0.54
1.2 Growth Performance			
Averaged BW starting (kg)	15.9±1.72 (n=6)	15.7±1.79 (n=6)	2.48
Averaged BW ending (kg)	23.0±1.04	19.0±1.49	1.82
Feed conversion rate (kg DM intake/kg gain)	6.63 <sup>b</sup> ±0.12	7.64 <sup>a</sup> ±0.07	0.62

<sup>1</sup> Standard error of difference, <sup>2 abc</sup>Values within the same column with different superscripts are significantly ( $P<0.05$ ) different. Values within the same column without different superscripts are not significantly ( $P<0.05$ ) different, *n*: numbers of animals, <sup>3</sup> using estimation model described by Jetana *et al.*, (2003)

### Effects of adding poly ethylene glycol 4000 (PEG) into rain tree pods as a feed supplement for Boer crossed bred goats on ruminal microbial N production

The experiment was conducted in six male *Boer* crossed bred goats (body weight ranges 29.5-39.0 kg). The animals were randomly divided into two groups of 3 animals each. All six animals were fed *ad libitum* with corn silages and supplemented either one of two supplemental pellets of RTPP (1.0 kg) and RTPP+PEG (1.0 kg RTPP+100 g PEG) in two different periods. The experimental design is crossing over (2 periods, each period 3 animals/treatment). Table 3 shows that the microbial N production in the rumen in goats were generally increased ( $P<0.05$ ) in the RTPP+PEG compared to those in the RTPP.

Table 3. Total OM intake and digestion and microbial N production in *Boer* crossed breeds goats fed corn silage and supplemented with rain tree pod pellets (RTPP) and rain tree pod pellets (RTPP) + polyethylene glycol

Dietary supplements	RTPP	RTPP+PEG	SED
Total DM intakes	96.0 <sup>b</sup> (n=6)	108 <sup>a</sup> (n=6)	4.62
Digestibility of OM	63.2 <sup>b</sup>	83.2 <sup>a</sup>	1.85
Total purine derivatives	1667 <sup>b</sup>	1922 <sup>a</sup>	105
PD (mmol)/kg DOMI	24.3 <sup>b</sup>	33.7 <sup>a</sup>	2.81
Microbial N (g N/day)	15.7 <sup>b</sup>	18.6 <sup>b</sup>	1.46

<sup>1</sup> Standard error of difference, <sup>2 abc</sup>Values within the same column with different superscripts are significantly ( $P<0.05$ ) different. Values within the same column without different superscripts are not significantly ( $P<0.05$ ) different, *n*: numbers of animals, <sup>3</sup> using estimation model described by Jetana *et al.*, (2003)

## Discussions and Conclusion

Feed represents the major cost for feed livestock production. Therefore, development of non-conventional, low-cost feed sources may reduce production costs, the high producing and fast growing

microbial populations in the rumen require mainly materials containing energy and protein in diets. In developing countries, livestock diets are based on agricultural by-products, which is general low quality. Therefore has been great interest in finding alternative sources of energy and protein to reduce livestock production cost. In this aspect, rain tree pods have attracted to the ruminant nutritionists due to high metabolizable/fermentable energy and high crude protein content. Though RTP can enhance microbial N production in the rumen but phenolic compounds (anti-nutritional agents, particularly condensed tannins) in RTP should be deactivated. In addition, RTP can be used as a source of energy that could replace molasses or other sugars extracted from fruits, vegetable or even in herbs that could help in reducing cost of feeding. This report provides an opportunity and a possibility of a great potential for developing rain tree pods as a functional feed and value-added feed for livestock production improvement.

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# Effect of Different Protein and Energy Levels in Concentrate Diets on Performances of Anglo-Nubian Goat During Late Pregnancy and Lactation

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## Abstract

*This trial was carried out to investigate the effects of different levels of protein and energy in concentrate diets on nutrient intake, live weights and feed conversion ratios of Anglo-Nubian goats during late pregnancy and lactation periods. Twelve young female Anglo-Nubian goats were divided into three groups and received different levels of crude protein (CP) and energy (as total digestible nutrient-TDN) in concentrate diets. The concentrate diets as follows: R1=16%CP, 64%TDN, R2=15% CP, 66%TDN and R3=14%CP, 68%TDN. Animals were offered King grass ad libitum, 1 kg of mixed forages and 1 kg of concentrate diets. The initial live weights were  $42.68 \pm 5.67$  kg. Feed intakes and live weights were measured daily and by weekly, correspondingly. The experiment was conducted in a completely randomized design with three treatments and four replications. The concentrate diets had no significant effects on nutrient intake, ADG and FCR ( $p > 0.05$ ) during late pregnancy. The mean of DM, TDN and CP intakes for all treatments were 1567, 1131 and 259 g/d, respectively; with the mean ADG and FCR were 209 g/d and 7.62. The concentrate diets had significant effects on DMI and TDN intakes and ADG ( $p < 0.05$ ) but had no effect on CP intake ( $p > 0.05$ ) during lactation periods. The DM and TDN intakes were 1760 and 1260 g/d, 1694 and 1219 g/d, and 1701 and 1220 g/d for R1, R2 and R3, respectively. The mean CP intake was 274 g/d for all treatments. The ADGs for R1, R2 and R3 were -45.71, -58.20 and -27.98 g/d, respectively. In conclusion, the level of protein and energy in concentrate diets did not affect the nutrient intake, ADG and FCR during late pregnancy but did affect the DMI and TDN intakes and ADG of Anglo-Nubian goats during lactation.*

*Keywords: Anglo-Nubian goats, energy, lactation, protein, pregnancy*

## Introduction

The Population of goat in Indonesia was around 19.22 million heads in 2014 (Livestock and Animal Health Statistics 2015) used for milk and meat production. To improve the quality and quantity of goat milk production, and an Anglo-Nubian goat was introduced in the breeding programme in the Indonesian Research Institute for Animal Production. This Anglo-Nubian breed often produces triplets as offspring (Mellado et al. 2011) and gives higher milk yield compared to Ettawa Grade (Praharani, 2014) and Angora goats (Anwar et al. 2015). Therefore, the Anglo-Nubian goats are used to produce a new breed of the goat with higher milk production and adapted to Indonesian environmental conditions.

Feeding goats involves combining various feedstuffs into an acceptable and palatable ration to meet nutrient requirements. These requirements vary depending on the stage of growth, gestation and lactation. The nutrients considered in diet formulation are energy, protein, minerals, vitamins and water. The balance of nutrients will determine the performance of a dairy goat. Pregnancy and delivery make demands on the does that should be met mostly from her diet. Lactation requires high levels of energy, protein, and water for milk production. The basal diets of dairy goats were often supplemented with the concentrate to meet their requirements.

An Anglo-Nubian goat is a newly introduced breed in Indonesia, the information on the feed intake, and nutrient utilization of this breed of goats under the traditional or intensive productions are scant in Indonesia. Supriyati et al. (2014) reported that levels of protein and energy in concentrates affected the total digestible nutrient (TDN) and crude protein (CP) intakes but did not influence the dry matter (DM) intake, average daily gain (ADG) and feed conversion ration (FCR) of young female Anglo-Nubian goats. The aim of the trial was to evaluate the effects of protein and energy at different levels in the concentrate diets on the performance of Anglo-Nubian goats during late pregnancy and lactation.

## Materials and Methods

Twelve young female late pregnancy Anglo-Nubian goats around two years of age with an average body weight of  $42.68 \pm 5.67$  kg were divided into three concentrate diets treatments. The treatments were formulated at different CP and TDN levels: R1= 16%CP, 64%TDN, R2=15% CP, 66%TDN and R3=14%CP, 68%TDN. Animals were offered King Grass *ad libitum*, 1 kg of mixed forages and 1 kg of concentrate diets. Table 1 shows the chemical composition of the feed. The experimental design was completely randomized in three treatments and four replications. Each animal was housed in individual cages. Cages had metal wire galvanized floors and attached to each cage was a secured woody container for feed. Water was provided through a nipple in each cage. Feed intakes were measured daily, and body weights were measured weekly. The treatments were carried out during late pregnancy (eight weeks at last gestation) and 12 weeks lactation period.

Parameters measured were DM, TDN and CP intakes, ADG and FCR. The DM, CP, neutral detergent fiber (NDF), acid detergent fiber (ADF), calcium (Ca) and phosphorus (P) contents of the grass, mixed forage and concentrate diets were analysed according to the AOAC method (AOAC 2005). Gross energy values were determined by bomb calorimeter (Adiabatic Bomb, Parr Instrument Co), and these values were used for TDN calculation as described by NRC (1981).

Table 1. Chemical composition of feed (%)

Variables	Grass	Mixed Forages	Concentrate diets		
			R1	R2	R3
Dry matter	29.38	40.56	88.94	88.23	87.78
Crude protein	9.09	20.56	16.21	15.28	14.33
Total digestible nutrient	67.20	74.44	64.15	66.14	68.01
Neutral detergent fiber	63.65	56.24	36.75	39.23	33.94
Acid detergent fiber	48.27	50.71	29.03	19.74	18.94
Calcium	0.34	1.39	0.91	1.05	0.99
Phosphorus	0.29	0.16	0.84	0.85	0.77

R1=16%CP 64%TDN, R2=15%CP 66%TDN, R3=14%CP 68%TDN

Feed intake, ADG and FCR of goats, were subjected to analysis of variance using General Linear Model (GLM) procedure of SAS (SAS Institute Inc. 2002). Differences between means were determined by Duncan's multiple range tests at a significance level of  $p < 0.05$ .

## Results and Discussion

Feeding of Anglo-Nubian goats with different levels of dietary protein and energy in concentrates had no effects ( $p > 0.05$ ) on the daily total DM, CP and TDN intakes, ADG and FCR among the treatment diets during late pregnancy (Table 2). The mean of daily DM, TDN and CP intakes for all treatments were 1567, 1131 (ME 10.87 MJ/kg) and 259 g/d, respectively. No differences in nutrient intake of feed goats resulted in no significant differences ( $p > 0.05$ ) on ADG and FCR during late pregnancy.

The mean daily TDN and CP intakes during late pregnancy in this trial were higher than the calculated requirement according to Langston University's Calculator (Langston University 2015). The requirement of TDN and CP for late pregnancy dairy goat of 50 kg BW and 210 g ADG were 1048 g and 204 g, respectively. Meanwhile, the requirement of DM, TDN and CP for goats 50 kg of BW and 120 g ADG were 1.43 kg, 1.00 kg and 153 g, correspondingly (Kearl 1982). Furthermore, Sahlou et al. (2004) reported that the metabolism of energy (ME) requirement for maintenance (ME<sub>m</sub>) was 462 kJ/kg BW<sup>0.75</sup> and for gain (ME<sub>g</sub>) was 28.5 kJ/g ADG for mature dairy goats. If the results of the trial calculated to the Sahlou's equation, the ME requirement in this experiment should be 14.64 MJ/kg for a dairy goat of 50 kg of BW and 209 g ADG. But the realized ME intake in this trial was 10.87 MJ/kg, 26% lower than Sahlou et al. (2004)'s equation.

The protein intake in this trial was also higher than the values of Sahlou et al. (1992)'s but lower than the values of Chaokaur et al. (2012)'s reports. Sahlou et al. (1992) reported that the protein requirement of dairy goats was 220 g/d during late pregnancy. Meanwhile, Chaokaur et al. (2012) reported that the protein requirement for growing Anglo-Nubian crossbred goats fed a *Leucaena leucocephalade* roughage-based diet with varying levels of cassava chips under tropical condition was 272 g/d.

Table 2 shows that the different levels of protein and energy in concentrate diets during lactation had significant ( $p < 0.05$ ) results in the daily DM intake, TDN intake and ADG. But the treatment diets had no effect ( $p > 0.05$ ) on CP intake during 12 weeks lactation. The DM and TDN intakes decreased with increasing the levels of dietary energy and decrease in the levels of protein. The mean daily DM and TDN intakes during lactation in this trial were lower for DM intake but higher for TDN and CP intakes than the values of Kears (1982)'s recommendation. The requirement of DM, TDN and CP intakes of goats 50kg BW and -20g ADG were 2.25kg, 1.15kg and 176g, correspondingly (Kears 1982). According to Langston University's Calculator, the requirements of DM and TDN and CP intakes were 1.70 kg, 1.03 kg and 164 g, respectively, for goats 50 kg of BW, -30g ADG, milk yield 1.2 kg, 5% fat and 3% protein.

The requirement of energy and protein during lactation in dairy goats had been reported by Sahlu et al. (2004) and Nsahlai et al. (2004). The dietary ME requirement for lactation was 5.224 MJ/kg 4% fat corrected milk, corresponding to an efficiency of ME use for lactation of 0.59. Furthermore, the metabolism protein (MP) requirement for lactation was 1.45 g/g milk protein, equivalent to a milk protein efficiency of 0.69 (Sahlu et al. 2004).

Table 2. Performances of goats fed different levels of protein and energy during late pregnancy and lactation

Variables	Treatment of concentrate diets			SEM	Significantly
	R1	R2	R3		
Late Pregnancy					
Total DM intake (g)	1570	1574	1557	9.67	NS
Crude protein intake (g)	256	268	256	8.55	NS
TDN intake (g)	1131	1140	1122	9.54	NS
ADG (g/h)	225	193	210	19.60	NS
FCR	7.07	8.23	7.55	0.72	NS
Lactation					
Total DM intake (g)	1760 <sup>a</sup>	1694 <sup>b</sup>	1701 <sup>b</sup>	20,00	S
TDN intake (g)	1260 <sup>a</sup>	1219 <sup>b</sup>	1220 <sup>b</sup>	15,00	S
Crude protein intake (g)	276	277	269	8,65	NS
ADG (g/h)	-45,71 <sup>b</sup>	-58.20 <sup>c</sup>	-27,98 <sup>a</sup>	6,41	S

R1=16%CP 64%TDN, R2=15%CP 66%TDN, R3=14%CP 68%TDN

SEM= standard error means, S=significant, NS=not significant

<sup>abc</sup>Value followed by different superscripts in the same row differ significantly ( $p < 0.05$ ).

The decrease of dietary protein by one % and increase TDN by two % in this trial gave no differences ( $p > 0.05$ ) on the ADG dan FCR between does on R1, R2, R3 diets during late pregnancy. As reported by Sahlu et al. (1992) body weight gains of Alpine goats increased with increasing CP intake from 164, 220, and 303 g/d. The prepartum ADG increased quadratically as protein (8.5, 11.5, and 14.5% of DM) amount increased but was unaffected by energy (50, 60 and 70% TDN of DM) on Alpine does (Sahlu et al. 1995). Increase of dietary energy as TDN by two % and decrease of dietary protein by one % in this trial gave differences ( $p < 0.05$ ) on the ADG between does on R1, R2, R3 diets during lactation. These changes in live weight might be due to the better response on the changing of energy than protein in diets. As Nascimento et al. (2014) reported that the supplementation with different levels of energy in TDN content of 65%, 75% and 85% at the same level of protein (20% CP) in dairy goats during lactation, promoted a positive effect, as increased productive parameters and reduced the number of days for the reestablishment of the reproductive parameters postpartum.

## Conclusion

The level of protein and energy in concentrate diets did not affect the nutrient intake, ADG and FCR during late pregnancy but did affect the DMI and TDN intakes and ADG of Anglo-Nubian goats during lactation.

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# Evaluation of Eleutherine (*Eleutherine americana*) as Feed Additive for Poultry

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## Abstract

The application of synthetic antibiotic has long been reported to produce negative effects on animal, since it has increasing microbiota resistance in digestive tract. In fact, the alternative antibiotic is urgently required to support animal production system. Therefore, a preliminary *in vitro* studies has been carried out to determine the potential of eleutherine as feed additive in poultry production. The bulbs of eleutherine were extracted using four different organic solvents: methanol, ethyl acetate, diethyl ether and hexane. The crude extracts were analyzed for their bioactive content, antioxidant activity using DPPH method and antibacterial activity using agar diffusion method. The results indicated that all four material extracts produced bioactive compounds of tannin. Average of antioxidant activity were 191.70, 115.70, 86.02 and 11.25  $\mu$ M AEAC for methanol, ethylacetate, diethylether and hexane, respectively. Furthermore, average inhibition growth for pathogen bacteria of *Escherichia coli* were 5.94, 4.75, 4.56 and 4.44 mm for methanol, ethylacetate, diethylether and hexane, respectively, while inhibition for *Staphylococcus aureus* were 5.37, 6.50, 5.37 and 5.18 mm for methanol, ethylacetate, diethylether and hexane, respectively. The presence of antioxidant and antibacterial activity could be attributed to the bioactive content of the extracts. It revealed that the bulbs of eleutherine extract was a potential source for feed additive as phytobiotic to replace synthetic antibiotics. Methanol was however the most potential solvent to be used for extracting the bulbs of eleutherine.

**Key words:** antioxidant, antibacterial, bioactive, eleutherine, extract

## Introduction

Feed additive, known as antibiotics, in the animal production system has long been applied to achieve a special purpose such as enhance growth and improve health status of animal. This feed additive is derived from plant or part of the plant such as leaves, seed or root used in animal feeding to improve animal performance (Windisch *et al.*, 2008). The use of this additive however has long been reported to produce negative effect on animal, as it is increasing microbiota resistance in the gastrointestinal of livestock (Windisch *et al.*, 2008), and also may create environmental problems of antibiotic residues. As a result, new additive of plant origin, considered to be natural products that the consumer would accept. However, complication arise of feed additives may vary widely with respect to botanical origin, processing material and composition of materials.

Eleutherine (*Eleutherine americana*) is plant that has been used as spice for asian food and herbal medicine (Ifesan *et al.*, 2010). Medical effect of eleutherine is attributed to its bioactive contents such as alkaloid, saponin, tripenoid, steroid, glikoside, tannin, fenolik and flavonoid. Ifesan *et al.* (2009; 2010) revealed that eleutherine extracts produce antibacterail effect on the gram-positive bacteria and few of gram-negative bacteria. It also has antifungis effect on specific fungus i.e *Aspegillus niger*, *Penicillium spp* and *Rhizopus spp* (Ifesan *et al.*, 2010). Additionally, elethuerine also has undigestible carbohydrate that produced positive effect on specific bacteria in the colon to improve health status (Gibson *et al.*, 2004). Recently, works by Phoem and Voravuthikunchai, (2013) reported that eleutherine extract could be used as prebiotic to stimulate the growth of nonpathogenic bacteria. Therefore, Phoem and Voravuthikunchai (2013) recommended to use eleutherine as functional food for human. The objectives of current study were investigate the potential of bulbs of eleutherine extract as fed additive (phytogenic/phytobiotic) sources for polutry feed through *in vitro* study of their antioxidant and antimicrobial activity as well as identification of their bioactive compounds.

## Materials and Methods

### Experimental Materials

The bulbs of eleutherine were purchased from local market. The bulbs were dried at room temperature until about 90% of dry matter and ground to pass through 1 mm screen. Total phenol, flavonoid and tannin

content of extract were analysed using a procedure of Senter *et al.* (1989). One part of bulbs sample was mixed throughly with two parts (w/v) of methanol, ethyl acetate, diethylether and hexane, respectively. The mixtures were shaking for 7 days at room temperature. The mixtures were filtered using whatman paper-screen to get a clear extract. Extracts were then rotary evaporated at 40°C to reduce volume and were air-dried to get pellet extracts. The individuals pellet were kept for bioactive compounds analysis, and antioxidant and antimicrobial activity evaluation.

### Evaluation of Antioxidant Activity

Method of DPPH (diphenylpicrylhydrazyl) assay was applied to evaluate antioxidant activity. Antioxidant activity was performing on the concentration of 0.25, 0.50, 0.75 and 1 mg of bulbs extract in the solution. The procedures of Krings and Berger (2001) suggested that measurement of free radical scavenging activity of the bulbs extracts is done by measurement of reduction in absorbance of methanolic DPPH solution at 517 nm of absorbance in the presence of the extracts. The initial concentration of DPPH was 0.1 mM and then the reading of absorbance was done after the solution stand for 30 min period. The samples were appropriately diluted, if absorbance decrease too much before 30 min period. The antioxidant activity was expressed as  $\mu\text{M}$  of AEAC (*ascorbic acid equivalent antioxidant capacity*).

### Evaluation of Antimicrobial Activity

Antimicrobial activity was determined using diffusion-agar method of Ayad *et al.*, (2000) on the selected bacteria of *E.coli* and *S. aureus*. An approximately  $10^8$  cfu of tested bacteria in 10 mL nutrient broth was incubated for 24 h at 37°C. 100  $\mu\text{L}$  of culture bacteria were added to 20 mL agar at 45°C on the dishes. The dish-agar was rounded 8 mm diameter with a steril pippete after solidification. Subsequently, 100  $\mu\text{L}$  of extracts in different concentration of extract materials i.e 0.25, 0.50, 0.75 and 1 mg of bulbs extract were dispensed in individual-dish wells. A synthetic antibiotics of tetracycline was used as positive control. The dishes were incubated for 24 h at 37°C and then the diameter of the inhibition zones (clear area) was measured.

### Experimental Design

A design of Block Randomized Design was applied, with 4 blocks (type of solvents) and 4 treatments within 3 or 4 replicates. The treatments were 0.25, 0.50, 0.75 and 1 mg of bulbs extract in the solution. Data were subjected to analysis based on the Steel and Torrie (1980) and Least Significant Different (LSD) was applied to analyse the treatment differences.

## Results and Discussion

### Chemical Composition

Profiles of bioactive components from bulbs of eleutherine, was extracting from methanol, ethylacetate, diethylether and hexane solvents are presented in Table 1. Surprisingly, the values in Table 1 indicated that not all solvents produced bioactive compounds. Bioactive concentration varied between type of solvents, in which ethylacetate had both flavonoid and tannin. This discrepancy is due to the lower sensivity of analysis used. Therefore, the concentration of pellet of bulbs eleutherine extract in the solution is not high enough to produce expected bioactive compound type.

Table 1. Bioactive composition of 1 mg pellet of crude extracted bulbs of eleutherine from methanol, ethylacetate, diethylether and hexane solvents

Solvents	Fenol (mg/kg)	Flavonoid (%)	Tannin (%)	Antioxidant ( $\mu\text{M}$ AEAC)
Methanol	Nd	nd	0.09	191.70
Ethylacetate	Nd	63.48	0.20	115.70
Diethylether	nd	nd	0.10	86.02
Hexane	nd	nd	0.04	11.25

Note : nd=not detected

### Antioxidant Activity

Antioxidant activity of bulbs of eleutherine extract is presented in Table 2. Generally, antioxidant activity is existing in the materilas, as it contains bioactive materials such as fenol, flavonoid and tanin. Antioxidant

activity of eleutherine extract varied amongst types of solvents, for which methanol produced the highest mean value of antioxidant activity (191.70  $\mu$ M AEAC) followed by 115.70, 86.02 and 11.25  $\mu$ M AEAC for ethylacetate, diethylether and hexane, respectively, for which hexane-extract is only producing antioxidant activity when its concentration is 1 mg in the solution. The values of antioxidant activity increases linearly as its concentration in the solution increases. This agrees with previous results of Rusdi *et al.* (2010; 2014) for methanol-extracted manggo seed and legume leaves, respectively. Furthermore, application of natural antioxidant in animal nutrition commonly produces a positive response in poultry performance. For instance, Radwan *et al.* (2008) found improvement in nutrients digestibility, feed efficiency, egg production and egg quality, when natural antioxidant was included in the hens diet. Moreover, inclusion of natural antioxidant during laying period significantly decreased melonaldehyde formation in egg yolk and had a positive effect on oxidative stability of shell egg and also improved fertility and hatchability. While, Abd El-Hakim *et al.* (2009) reported that antioxidant function of plants or plant-extracts improved significantly live weight gain at the first 3 weeks of age and this trend is not significant at the 42 days of age. Additionally, extracted materials from plant generally improved broiler performance (Hernandez *et al.*, 2004; Aengwanich *et al.*, 2009). Extracts from seed coat of tamarind were used as antibiotic replacement to which improved broiler liveweight gain but did not influence the feed intake and feed conversion (Aengwanich *et al.*, 2009).

Table 2. Antioxidant activity of bulbs extracts from methanol, ethylacetate, diethylether and hexane at the level 0.25, 0.50, 0.75 and 1 mg of extracted material in the solution (n=3)

Solvents	$\mu$ M of AEAC				SEM
	0.25	0.50	0.75	1.0	
Methanol	105.52 <sup>a</sup>	142.22 <sup>b</sup>	204.45 <sup>c</sup>	314.55 <sup>d</sup>	23.90
Ethylacetate	27.22 <sup>a</sup>	77.67 <sup>b</sup>	141.20 <sup>c</sup>	216.81 <sup>d</sup>	21.60
Diethylether	68.66 <sup>a</sup>	76.29 <sup>b</sup>	83.59 <sup>c</sup>	115.53 <sup>d</sup>	5.40
Hexane	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	45 <sup>b</sup>	6.10

Note : Different superscript letters on the row and mean value of solvents (column) are significantly different (P<0.01)

### Antimicrobial Activity

Antimicrobial activity of bulbs of eleutherine from methanol, ethylacetate, diethylether dan hexane for pathogenic bacteria of *Escherichia coli* and *Staphylococcus aureus* is summarized in Table 3. Antimicrobial activity of current study agrees with the previous results of Akiyama *et al.* (2001); Pereira *et al.* (2007); Vijayalakshmi *et al.* (2011) and Sakunpak and Panichayupakaranant (2012) who demonstrated the antimicrobial activity of polyphenol substances (such as tannin) from extracted plant. Polyphenol has ability to control microfauna and microflora population in the digestive tract of animal. Additionally, polyphenol, phenol, flavonoid and essential oil are extracted materials reduced the growth of pathogenic bacterial of *E. coli*, *S.aureus*, *L. monocutogenes* and *Salmonella spp* (Friedman *et al.*, 2004; Oussalah *et al.*, 2006). The results of present study also revealed that antibacterial activity is dosis dependant, for which antibacterial activity was firstly existing when the concentration of the extract is 0.5 mg (Table 3). The activity tended to increases as concentration of crude extract increases in the solution. The existence of antibacterial activity for both gram-positive and gram-negative bacteria revealed that the extract of bulbs eleutherine, in the present study, is potentially to be used as a wide broad spectrum antibiotics.

Table 3. Inhibition bacterial growth of bulbs extract from different solvents on *Escherichia coli* and *Staphylococcus aureus* at the level 0.25, 0.50, 0.75 and 1 mg of extracted material in the solution (n=4)

Solvents	Inhibition (mm)									
	<i>Escherichia coli</i>					<i>Staphylococcus aureus</i>				
	0.25	0.50	0.75	1.00	SEM	0.25	0.50	0.75	1.00	SEM
Methanol	nd	3.00 <sup>a</sup>	7.50 <sup>b</sup>	13.25 <sup>c</sup>	1.30	nd	4.00 <sup>a</sup>	7.25 <sup>b</sup>	10.25 <sup>c</sup>	0.99
Ethylacetate	nd	3.00 <sup>a</sup>	7.75 <sup>b</sup>	8.25 <sup>c</sup>	0.89	nd	7.25 <sup>a</sup>	8.75 <sup>b</sup>	10.00 <sup>c</sup>	1.01
Diethylether	nd	3.75 <sup>a</sup>	6.00 <sup>b</sup>	8.50 <sup>c</sup>	0.82	nd	4.75 <sup>a</sup>	6.00 <sup>b</sup>	10.75 <sup>c</sup>	1.01
Hexane	nd	4.25 <sup>a</sup>	5.75 <sup>b</sup>	7.75 <sup>c</sup>	0.75	nd	5.50 <sup>a</sup>	8.50 <sup>b</sup>	9.25 <sup>c</sup>	0.95
Tetracycline			26.00					25.50		

Note : nd =not detected. Different superscript letters on the row within type of bacteria are significantly different (P<0.01)

Bioactive compounds in the particular medium generally produce activity as antioxidant and antibacterial on particular bacteria and fungus, even more may reduce the growth of mosquito larvae (Ferreira *et al.*, 2008). The absence of antibacterial activity on lower concentration of extract in the solution could be explained that bioactive content in the solution is not high enough to inhibit the growth of tested bacteria. Previous results of Banso and Adeyemo (2007); Vijayalakshmi *et al.* (2011) and reported that minimum inhibitory bioactive concentration of tannin ranged from 4.0 to 5.5 mg/mL. While, Sakunpak and Panichayupakaranant (2012) found a value of 10 mg/mL to produce antibacterial activity. Additionally, low activity compounds had a high concentration to produce antibacterial activity, while high activity agent gave a low minimum concentration (Banso and Adeyemo, 2007).

## Conclusion

Extracts from bulbs of eleutherine in the solvents of methanol, ethylacetate, diethylether and hexane produced antibacterial and antioxidant activity. Therefore, bulbs of eleutherine is potential to be used as feed additive of phytobiotic. Methanol solvent was however the most potential solvent to be used to extract materials from bulbs of eleutherine.

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# The Effect of NaOH Concentrations and Polysaccharides Extract of Palm Kernel Meal on Performance of 4 Weeks Old-Broiler Chickens

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## Abstract

NaOH has been used as a solvent to extract polysaccharides, particularly in the product of *palmae* species. The polysaccharides produced was mainly mannose – based that is potential to be used as a prebiotic. A study was carried out to determine the effect of level of NaOH concentration and polysaccharides extract of palm kernel meal on performance of 4 weeks old-broiler chickens. A total of 120 unsexed broiler chicks was used in this study. The broiler chicks were kept in cages for 4 weeks. The birds were fed with starter diets for the first three weeks and grower diets from 3 weeks to 6 weeks. Feed and water were offered ad-libitum. The experimental diets were three levels of NaOH concentration when extracting the palm kernel meal polysaccharides (10, 20 and 30% NaOH) and two levels of the polysaccharides extract (0,025 and 0,05% in the diet). A completely randomised factorial design was used with three levels of NaOH concentration for extracting the polysaccharides from palm kernel meal and 2 levels of palm kernel meal polysaccharides and four replicate cages. Differences found among treatments were tested with Tukey test. The results indicated that concentration of NaOH used for extraction process did not statistically produce any significant difference in body weight gain, feed intake and FCR when fed to broilers kept for 4 weeks. The effect of polysaccharides extract was significant in FCR, but not in body weight gain and feed intake. Interaction between NaOH concentration and polysaccharides extract of palm kernel meal was found in feed intake and FCR in which the increased concentration of NaOH up to 30% produced higher feed intake when the birds fed 0.025% polysaccharides extract of palm kernel meal. FCR was decreased when the diet was supplemented with 0.025% palm kernel polysaccharides in the palm kernel meal polysaccharides extracted with 10% NaOH, not in 20 and 30% NaOH. In conclusion, FCR was affected by polysaccharides extract and interaction between concentrations of NaOH and polysaccharides extract of palm kernel meal affected feed intake.

Keywords: Palm kernel meal, mannose based polysaccharides, broiler.

## Introduction

The major ingredient of carbohydrates in palm kernel meal has been reported to be mannose based polysaccharides (Duesterhoft et al., 1991; Daud and Jarvis, 2002). The use of NaOH to extract mannose based polysaccharides from palm nuts has been reviewed by Sundu et al. (2006). Considerable amount of mannose based polysaccharides (mannan) was produced when adopting the method of Kusakabe and Takashi (1988), who used NaOH as a solvent to extract polysaccharides. Since mannose based polysaccharides from yeast (*Saccharomyces cereviceae*) has been used as a prebiotic to promote growth of broiler chickens, polysaccharides from palm kernel meal may also play the same role found in yeast mannan.

A study of using copra mannan conducted by Sundu and Damry (2008) indicated that copra mannan behave like yeast mannan either in binding pathogenic bacteria in the digestive tract of broiler chickens or binding mycotoksins (Sundu et al., 2006). Since coconut tree and oil palm tree are classified in the same family, palm kernel polysaccharides may have the same property as coconut polysaccharides and thus is potential to be used as prebiotic. Fernandez et al. (2002) indicated that palm kernel meal could decrease the pathogenic bacteria population in the digestive tract of broilers. Accordingly, a study of the use of palm kernel polysaccharides extracted with different concentrations of NaOH and levels of palm kernel polysaccharides in the diets was conducted to determine their effects on broiler performance.

## Materials and Methods

### Mannan extraction

The extraction of palm kernel polysaccharides was done by adopting the method of Kusakabe and Takashi (1988). Different concentrations (10%, 20% and 30%) of NaOH were used as solvent, with the ratio

16 l NaOH to 2 kg of palm kernel meal (PKM). The mixture of PKM and NaOH was occasionally stirred and left for 24 hours at room temperature. The mixture was filtered through a cloth bag. The filtrate was neutralized with 12 N H<sub>2</sub>SO<sub>4</sub>. Resultants precipitate (mannose based polysaccharides) were collected after dialysing against tap water to remove salts. The leftover residue was stored as palm kernel polysaccharides.

### Birds and Feed

This study was conducted in the poultry station, Faculty of Animal Husbandry and Fisheries, the University of Tadulako Palu, Indonesia. A total of 120 day old unsexed broiler chicks was used. The birds were placed in brooder cages for two weeks. On day 14, the broiler chickens were transferred into 24 cages. From day 1-21, the broiler chicks were fed starter diets and grower diets (Table 1) from day 22 - 42. The birds were fed basal grower diet (Table 1) *ad-libitum* and water was available throughout the trial and treatment diets were in Table 2.

Table 1. Composition of the experimental control basal diet (%)

Ingredients	Starter diet	Grower diet
Full fat soybean meal	24.99	18.97
Corn	60.20	62.10
Fish meal	11.20	11.00
Rice bran	2.0	3.9
Palm oil	0.38	1.0
Dicalcium phosphate	1.30	1.2
Salt	0.07	0.2
Methionine	0.15	0.15
Lysine	0.05	0.11
Vitamine and Mineral Mixture	0.20	0.20
Calculated :		
Crude protein	23.13	21.00
Crude fibre	3.5	3.6
ME (K Cal/kg)	3200	3187
Lysine	1.1	1.0
Methionine	0.4	0.4
Calcium	1.4	1.0
Phosporous	0.9	0.7

Table 2. Treatment diets

Extracted polysaccharides	Levels in the diet	Replications	Birds
10%	0.025%	4	5
	0.05%	4	5
20%	0.025%	4	5
	0.05%	4	5
30%	0.025%	4	5
	0.05%	4	5

### Statistical analysis

This trial used a completely randomized factorial design with three levels NaOH concentrations when extracted the polysaccharides, 2 levels of polysaccharides in the diets and four replicate cages of five birds each cage. Data were analysed by analysis of variance and differences among treatments were tested by TukeyTest (Steel and Torrie, 1980).

### Results and Discussions

The data on the effect of various concentrations of NaOH to extract palm kernel polysaccharides were shown in Table 3. The effect of levels of polysaccharides in the diet can be seen in Table 4 and their interaction are presented in Table 5. There were no significant effect ( $P>0.05$ ) of levels of NaOH concentration on body weight gain, feed intake and feed conversion ratio (FCR). Levels of polysaccharides in the diets produced significant effect ( $P<0.05$ ) only on FCR. Interaction was found in feed intake and FCR.

Table 3. The effect of palm kernel polysaccharides extracted with different NaOH concentrations on body weight gain, feed intake and FCR

Treatments	Body weight gain (g)	Feed intake (g)	FCR
PKM extracted with 10% NaOH (T1)	1063 <sup>a</sup>	1781 <sup>a</sup>	1.69 <sup>a</sup>
PKM extracted with 20% NaOH (T2)	1062 <sup>a</sup>	1787 <sup>a</sup>	1.69 <sup>a</sup>
PKM extracted with 30% NaOH (T3)	1094 <sup>a</sup>	1881 <sup>a</sup>	1.72 <sup>a</sup>

Table 4. The effect of levels of Palm kernel polysaccharides in the diets on body weight gain, feed intake and FCR

Polysaccharides in the diet	Body weight gain (g)	Feed intake (g)	FCR
0.025% in the diet (P1)	1103 <sup>a</sup>	1821 <sup>a</sup>	1.65 <sup>b</sup>
0.05% in the diet (P2)	1043 <sup>a</sup>	1811 <sup>a</sup>	1.75 <sup>a</sup>

Table 5. Effects of interaction between NaOH concentration and palm kernel polysaccharides levels in the diets on broiler performance at 4 week of age

Treatments	Feed intake (g)		Treatments	FCR	
	P1	P2		P1	P2
T1	1648 <sup>a</sup> <sub>q</sub>	1913 <sup>a</sup> <sub>p</sub>	T1	1.58 <sup>b</sup> <sub>p</sub>	1.79 <sup>a</sup> <sub>p</sub>
T2	1876 <sup>a</sup> <sub>pq</sub>	1698 <sup>a</sup> <sub>p</sub>	T2	1.69 <sup>a</sup> <sub>p</sub>	1.70 <sup>a</sup> <sub>p</sub>
T3	1939 <sup>a</sup> <sub>p</sub>	1823 <sup>a</sup> <sub>p</sub>	T3	1.70 <sup>a</sup> <sub>p</sub>	1.75 <sup>a</sup> <sub>p</sub>

Different superscripts (ab) within rows mean significantly different ( $P < 0.05$ )

Different subscripts (pq) within column mean significantly different ( $P < 0.05$ )

Body weight gain, feed intake and FCR were not affected by levels of NaOH when extracted palm kernel polysaccharides. An improved FCR was found in the birds fed 0.025% polysaccharides. This indicated that the birds fed 0.025% palm kernel polysaccharides were more efficient in converting feeds into bodyweight gain. Increased palm kernel polysaccharides inclusion in the diet decreased the quality of the diet.

Interaction was found in feed intake and FCR but not in body weight gain. Increased levels of palm kernel polysaccharides in the diets did not affect feed intake of birds fed the diets containing polysaccharides extracted with various concentrations of NaOH (10%, 20% and 30%). However, feed intake increased significantly as the level of NaOH concentration as solvent increased in the diets supplemented with 0.025% palm kernel polysaccharides, but not in the diets containing 0.05% palm kernel polysaccharides. It can be speculated here that the use of the lowest concentration of NaOH as solvent was more efficient in the diets supplemented with 0.25% palm kernel polysaccharides.

Feed conversion ratio was not affected when the broiler chickens were fed palm kernel polysaccharides extracted with different levels of NaOH concentration. Interestingly, lowering the inclusion level of palm kernel meal polysaccharides improved FCR in the diets extracted with 10% NaOH, but not in the diets extracted with 20 and 30% NaOH. Since the NaOH as a solvent needs to be minimally used due to the fact the residual fluid of the extraction process could impair the environment, these findings produced promising results as the lowest concentration of NaOH and the lowest polysaccharides inclusion gained better performance. In conclusion, the birds fed the palm kernel polysaccharides extracted with the lowest concentration of NaOH (10%) and the lowest inclusion of palm kernel meal polysaccharides (0.025%) produced good result.

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# Combination Effect of Nutritech Feed Additive Containing Saponin, Tanin and Eugenol Essential Oils on *in Vivo* Rumen Methane Production in Dairy Cattle Using Open Circuit Respiration Chamber Technique

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## Abstract

Diet manipulation containing feed additive for reducing enteric methane emission of ruminant had been an interesting focus since the GHG's emission of livestock was reported increasing. Saponin, tannin and eugenol had been selected when they have a good effect for rumen methane gas reduction. There is no more information for combination between saponin, tannin and eugenol which was tested for rumen methane reduction, especially in vivo dairy cattle methane gas production assessment. Therefore, this study was carried out to evaluate the effect of combination of saponin-containing *H. Rosa sinensis* leaf, tannin-containing tea waste and eugenol-containing clove oil on in vivo rumen methane production assessment in dairy cattle using open circuit respiration chamber technique. The combination was mixed in one feed additive, Nutritech-BPPT. The treatments were R1: control ration (60% forage + 40% concentrate), R2: R1 + nutritech 100 mg/kg of body weight, R3: R1 + nutritech 150 mg/kg of body weight and R4: R1 + nutritech 200 mg/kg of body weight, which were assigned and analyzed in latin square design (4x4). Four dairy cows and four heifers Fries Holland (PFH) were used for in vivo rumen methane production assessment using open respiration chamber technique (MARS-Sable System International). The results indicated that supplementation of nutritech-BPPT reduce methane production from dairy cattle 14.96%, 25.41% and 34.92% after supplementation 200 mg/Kg BW, 100 mg/Kg BW and 150 mg/Kg BW of nutritech, respectively. The combination between saponin, tannin and eugenol essential oil in Nutritech was formulated as an optimum composition after in vitro test and their effect revealed that saponin, tannin and clove oils were no effect on in vitro rumen digestibility. Clove essential oil also may have strong microbial activity from their active component (eugenol). Eugenol is main component of clove oil which its composition was very high more than 90% in clove oil for this study. In conclusion, Nutritech containing saponin, tannin and eugenol essential oil could be used as feed additive for rumen methane reduction in dairy cattle.

**Keyword:** dairy, eugenol, methane, saponin, tannin

## Introduction

Diet manipulation containing feed additive for reducing enteric methane emission of ruminant had been an interesting focus since the GHG's emission of livestock was reported increasing. Saponin, tannin and eugenol had been selected when they have a good effect for rumen methane gas reduction. There is no more information for combination between saponin, tannin and eugenol which was tested for rumen methane reduction, especially in vivo dairy cattle methane gas production assessment. Therefore, this study was carried out to evaluate the effect of combination of saponin-containing *H. Rosa sinensis* leaf, tannin-containing tea waste and eugenol-containing clove oil on in vivo rumen methane production assessment in dairy cattle using open circuit respiration chamber technique

Open circuit respiration chamber technique is one method for determine methane emission from ruminant. In the open-circuit respirometry chamber, outside air is supplied to the chamber continuously and chamber air is removed (Soliva and Hess, 2007). Measuring CH<sub>4</sub> emission from enteric fermentation using open-circuit respirometry chamber was reported has similar values Dry matter intake (DMI) and Methane production by grazing cattle to those obtained using a micrometeorological dispersion model (Tomkins et al., 2011). Rumen modification strategies by diet manipulation using supplementation oils and plant secondary active compound also has been assessed by some researcher (Patra, et al., 2011; calsamiglia et al., 2007 and Gorgulu et al., 2010). Their report was variation between animal, plant bioactive compounds, measurement technique and doses.

Eugenol is a member of the [phenylpropanoids](#) class of chemical compounds as a major component of clove oils. Tannins is major component of tea and saponin is a major component of *Hibiscus rosasinensis* leaf plant. They are abundant available in Indonesia and could be effectively is used as feed additive ingredient for ruminant. Their capacity in reducing methane from ruminant was reported well. Combination strategies between some plant bioactive may result in additive and/or synergistic effects that may enhance efficiency of rumen microbial fermentation and nutrient utilization in ruminants. There was antagonistic effect of combination between clove oil and orange peel oil 1.8 g/d on in vivo and in vitro rumen methane production (ml/gDM and g/DMI) (Rofiq et al., 2014) but synergic effect of combination between clove oil and cinnamaldehyde. Hence, their doses for combination is required in rumen methane reduction and feed efficiency. There is no more information for combination between saponin, tannin and eugenol which was tested for rumen methane reduction, especially in vivo dairy cattle methane gas production assessment. Therefore, this study was carried out to evaluate the effect of combination of saponin-containing *H. Rosa sinensis* leaf, tannin-containing tea waste and eugenol-containing clove oil on in vivo rumen methane production assessment in dairy cattle using open circuit respiration chamber technique. Nutritech-BPPT is a feed additive containing saponin, tannin and eugenol feed Additive containing saponin, tannin and eugenol.

## Materials and Methods

Four heifers Fries Holland (PFH) were used for in vivo rumen methane production assessment using open respiration chamber technique (MARS-Sable System International). They fed total mixed ration as a control ration (60% forage + 40% concentrate) with Nutritech-BPPT feed additive as a treatment for this experiment. Nutritech-BPPT is a feed additive containing 6% saponin, 40% tannin and 0.8 % eugenol of nutritech. The treatments were R1: control ration (60% forage + 40% concentrate), R2: R1 + nutritech 100 mg/kg of body weight, R3: R1 + nutritech 150 mg/kg of body weight and R4: R1+ nutritech 200 mg/kg of body weight, which were assigned and analyzed in latin square design (4x4).

Rumen methane production from heifers were measured by open respiration chamber technique. After treatments with Nutritech supplementation, animals were located in open circuit respiratory chamber for 5 days (3 days for chamber adaptation and 2 days respiration measurements). Respiration measurements using MARS Sable System USA<sup>(R)</sup>, an open circuit respiratory system for measuring gas containing in gas respiration, pressure and flow. Chamber with close head animal keep gas respiration near animals head that would took by pump flow meter via plumbing. The gas from flow meter is filtered into the scrubber and gas dryer before entre to gas analyser. There are 2 chambers for 2 treatments which were automatically arranged by Intelligent Multiplexer (RM8) for changing measurements between chamber and baseline measurement. The gas analyser reads gas contains (CH<sub>4</sub>, O<sub>2</sub>, and CO<sub>2</sub>) as percentage gas composition. It would be read by computer with acquisition program Expedata Software and the result of gas contains could be reported with graph and data sheet.

## Results and Discussion

Result showed that supplementation of nutritech-BPPT reduce methane production from dairy cattle 25.41%, 34.92% and 14.96% after supplementation 100 mg/Kg BW, 150 mg/Kg BW and 200 mg/Kg BW of nutritech, respectively (Table 1). Combination effect between tannin, eugenol and saponine was effective at 150 mg doses of Nutritech.

Supplementation 100 mg of Nutritech Combination effect might give specific function of tannin, eugenol and saponine. Tannin and saponin are reported could reduce rumen protozoa and reduce methanogen bacteria, indirectly. Eugenol has strong antimicrobial could reduce some microbes inside rumen. Eugenol reduced rumen pH and Methane production (Patra, 2009), N-NH<sub>3</sub> (Bach et al., 2005), increased propionic acid (Kung et al., 2008) and reduced butyric and acetic acid. Eugenol also reported that methane reducing 60-70% by feeding with clove oils (Patra, 2009). Its capability for decreasing methane is still need further research with some doses.

Table 1. Methane production of dairy cattle with Nutritech supplementation

Parameters	R1	R2	R3	R4
Dry matter intake				
DMI (Kg/d)	4.20 ± 0.26	4.12 ± 0.36	4.12 ± 0.15	4.07 ± 0.41
DMI (g/Kg BW)	182.19 ± 35.90	243.53 ± 53.59	279.92 ± 68.71	212.15 ± 51.09
Methan as measured				
CH <sub>4</sub> (L/d/head)	155.02 ± 24.51 <sup>c</sup>	114.06 ± 31.38 <sup>b</sup>	99.17 ± 24.97 <sup>a</sup>	128.03 ± 33.97 <sup>b</sup>
CH <sub>4</sub> (g/d/head)	99.37 ± 15.71 <sup>c</sup>	73.11 ± 21.12 <sup>b</sup>	63.57 ± 16.01 <sup>a</sup>	82.07 ± 21.77 <sup>b</sup>
CH <sub>4</sub> (g/kg DMI)	23.68 ± 45.08 <sup>c</sup>	17.66 ± 4.49 <sup>b</sup>	15.41 ± 3.77 <sup>a</sup>	20.13 ± 5.27 <sup>b</sup>
Methane predicted				
CH <sub>4</sub> (g/d) <sup>1</sup>	114.57 ± 1.60	113.52 ± 2.25	113.44 ± 0.94	112.85 ± 2.55
CH <sub>4</sub> (g/d) <sup>2</sup>	116.01 ± 4.21	113.24 ± 5.92	113.02 ± 2.48	111.47 ± 6.71

R1 = Control Feed (60% Grass 40% concentrated Feed), R2 = R1+100 mg/Kg BW, R3 = R1+150 mg Nutritech/kg BW, R4 = R1+400 mg Nutritech/Kg BW, DMI = dry matter intake, CH<sub>4</sub>(g/d)<sup>1</sup> =Ellis et al (2007), CH<sub>4</sub>(g/d)<sup>2</sup> =Kurihara (1999)

Compare value of methane production between methane as measured with methane prediction was not similar. Methane production estimation was higher than methane production as open respirometry chamber measurement. The estimation used dry matter intake of ruminant with combine database of beef and dairy database (Ellis, 2007) was over estimation for open respirometry chamber technique.

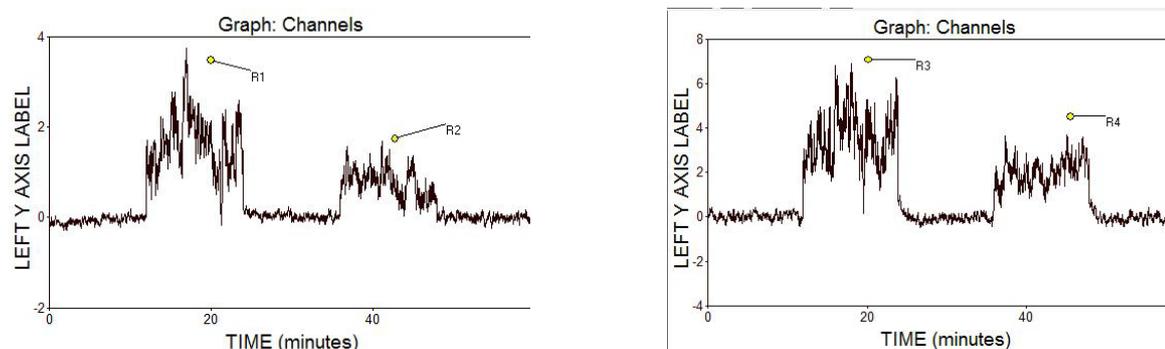


Figure 1. Methane production graph using Sable system MARS respiration measurement

## Conclusion

Supplementation of Nutritech containing saponin, tannin and eugenol could reduce methane by dairy cattle. Encapsulation of Nutritech as supplement for reduce methane by dairy goat could be used in future experiments.

## Acknowledgement

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# Growth and Feed Efficiency of Male Lambs Fed on Grass or Enriched Corn Cob Silage Basal Diet

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## Abstract

Ground corn cob is one agricultural by-products which potential to be used as fiber sources in ruminants ration to overcome forage scarcity during dry season. Study was conducted to evaluate growth and feed efficiency of male lambs fed on elephant grass or enriched corn cob basal diet. The study used 15 heads post weaning male lambs placed in individual pen last for 10 weeks. The lambs were grouped into 5 according to their body weight, the initial body weight was  $16.24 \pm 2.97$ g. Each animal in each groups were randomized to be allocated in one of three diet treatments which was different of basal diets, consisted of elephant grass (EG); corn cob silage enriched by corn grain (SC) and corn cob silage enriched by molasses (SM). The diet consisted of basal diet and concentrate with ratio 40:60%. The diets were formulated in iso energetic and iso protein to contain 14% CP and 2.5 Mcal/kg EM. Ground corn cob was mixed either with corn grain powder or molasses at 1% of DM corn cob at the beginning of making silage. Parameter recorded were average daily gain (ADG), dry matter intake (DMI), and feed conversion ratio. Results of the study shows that lambs fed on different basal diet their average daily gain was not significantly different with the ADG was 110.0; 102.7 and 97.7 g/day respectively for diet EG, SM and SC; similarly feed conversion ratio was also not significantly different with values 6.55; 87.6 and 8.49 respectively for diet EG, SM and SC. From this study it can be concluded that in iso energetic and iso protein diets, corn cob silage enriched either by corn grain or molasses can be used as basal diet for grass basal diet replacement.

**Keywords:** corn cob silage, feed efficiency, growth, lambs.

## Introduction

Corn is one of food crops that targeted by Indonesian to domestically self sufficient, many programs have been done to achieve this program. The increase of production is followed by the increase of by product such as corn stover, dried corn husk and corn cob. Except con corn, maize by products in Indonesia commonly used as large ruminants feed. This corn cob is potential to be used as fiber source in ruminants feed to overcome the forage scarcity during dry season. However, there is limitation of using corn cob as feed source. Besides its low quality, in long duration of storage, corn cob is prone to be contaminated by *Aspergillus flavus* which is very toxic. Ensiling is one of methods which can be used for preservation during storage and also can improve the palatability of feed. Quality of silage is affected by the availability of readily available carbohydrate (RAC) which can be used by lactic acids bacteria to be converted into lactic acid (McDonald et al., 2002). The low quality of corn cob was indicated by the high content of cell wall (NDF) (Yulistiani *et al.* 2012) so that the availability of RAC in corn cob is limited. To obtain good quality silage, the feeds must contain sufficient readily available carbohydrate so that the lactic acid bacteria able to convert RAC into lactic acid so that acid condition of silage can be obtained. Molasses and ground corn are available abundantly in Indonesia and can be used as readily available carbohydrate sources in making silage. In Indonesia lack of information of using corn cob silage with different RAC as basal diet for ruminants therefore the objective of this study was to evaluate the effect of fully replacement of grass basal diet with corn cob silage either enriched with molasses or corn powder on the growth of lambs

## Materials and Methods

The study used 15 head of post weaning male lamb with the average body weight of  $16.24 \text{ kg} \pm 2.975$ kg the lamb were grouped into five groups based on their body weight and placed in individual pen during 3 months of growth trial study. Each lambs in each group was randomly allocated to one of three diet treatments. The treatment diets was a different of basal diet as follows: 1. Grass Basal diet (EG); 2.

Corn cob silage enriched with ground corn (SC) basal diet. 3. Corn cob silage enriched with molasses (SM) basal diet; The ration consisted of basal diet and concentrate in ratio 40:60% and was offered in total mixed ration and were offered ad libitum. The ration was formulated in iso protein and iso energetic contained CP 14% and EM 9 MJ/kg. Concentrate were formulated using feed ingredients of coconut meal, rice bran, ground corn grain, soybean meal, molasses, mineral mixed, urea and salt. Silage was made by mixing ground corn cob with ground corn (SC) or Molasses (SM) as source of RAC at 2% of DM corn cob, after that the mixture of ground corn cob and RAC was mixed with water to obtained dry matter mixture of 30-40%. The mixture then filled into plastic bag and kept in anaerob condition for 21 days. The chemical composition of 3 basal diets is presented in Table 1. Feed offered and feed refusal were measured daily to obtained feed consumption data. While average daily gain were obtained by weighing the lambs weekly. Study was conducted in randomized complete block design. Data were analyzed by ANOVA using General Linear Model procedure (GLM) of SAS 6.12 (1989). Differences among treatment means were detected using Duncan's multiple range tests. Significance was taken at  $P < 0.05$ .

Table 1. Chemical composition and in vitro digestibility of Elephant grass, corn cob silage enriched by molasses and corn cob silage enriched by ground corn used as basal diet in the experiment

Variable	Elephant Grass	SM	SC
Organic matter (%)	89.4	98	98
Crude Protein (%)	6.56	3.51	4.8
Neutral detergent fiber (%)	68.32	79.98	75.78
Acid detergent fiber (%)	48.67	42.7	43.94
Lignin (%)	7.35	5.98	5.71
In Vitro dry matter digestibility (%)	54.68	51.94	46.5
In Vitro organic matter digestibility (%)	51.51	51.2	48.76

SM: corn cob silage enriched with molasses; SC: corn cob silage enriched with ground corn

## Results and Discussion

Feed consumption, average daily gain (ADG) and feed conversion is presented in Table 2. From table 2 can be seen that total dry matter intake (DMI) was not affected by different basal diet. Total DMI and %DMI/BW were similar this indicate that ensiling was able to increased palatability of corn cob so that the consumption of corn cob silage based diet comparable to grass basal diet. Previously Yulistiani *et al.* (2012) also reported that consumption corn cob silage based diet was comparable to fresh chopped elephant grass basal diet. Similar to DMI intake, the average daily gain was not significantly different between treatment as a results of the sheep obtained similar nutrition from the diet. This indicated in iso energetic and iso protein diet, different basal diets were able to produce similar performance, though corn cob silage either enriched by molasses (SM) or ground corn grain (SC) have lower nutrition than elephant grass as shown in Table 1. From Table 1 shows that SM or SC had lower protein content. The feed conversion ratio was also not significantly different between treatment this was due to the the sheep had similar feed consumption and similar ADG. It shows that these three diets have similar feed efficiency. From feed consumption, growth response and feed conversion it shows that feeding corn cob silage either enriched with molasses or ground corn cob produced similar response to grass basal diet therefore from this study the lack of grass forage in dry season could be replaced by corn cob silage as basal diet.

Table 2. Dry matter intake (DMI), average daily gain and feed conversion ratio of sheep fed on different basal diet

Variable	Basal diets			SEM
	EG	SM	SC	
Total DMI intake (g/day)	785.4	898.4	721.1	57.765
%DMI/BW	3.70	3.95	3.99	0.0988
Average daily gain (g/h/d)	110	102.7	97.7	5.9766
Feed conversion ratio	6.55	8.76	8.49	0.6722

EG: elephant grass; SM, corn cob silage enriched with molasses; SC, corn cob silage enriched with ground corn grain.

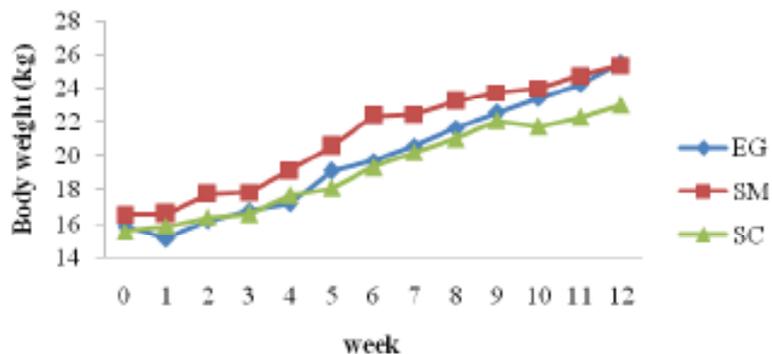


Figure 1. Body weight of lambs fed on different basal diet during growth trial period.

Note: EG, elephant grass; SM, corn cob silage enriched with molasses; SC, corn cob silage enriched with ground corn grain.

The body weight of lambs during feeding trial is shown in Figure 1. This figure shows that lambs still in growth stage as indicated by linear trend of the graph. Sheep fed on SC their body weight was constantly increased during the experiment but at 9<sup>th</sup> week of experiment in SC diet the sheep experience drop in body weight this might caused the lower ADG of SC (Table 2). SM and EG basal diet on the other hand, their body weight decreased at early experiment but after adjustment, their body weight costantly increased.

## Conclusion

From this study can be concluded that corn cob silage either enriched with molasses or ground corn in iso energetic and iso protein ration could fully replaced fresh grass basal diet without any negative effect on lamb growth rate dan produced similar feed efficiency.

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# Nitrogen Utilization and Ruminal Fermentation of Five Breed of Sheep Fed Concentrate Containing Different Levels of Rumen Undegradable Protein

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## Abstract

Study was conducted to evaluate the effect of feeding different levels of rumen undegradable protein (UDP) in concentrate from feed ingredients on nitrogen utilization and ruminal fermentation of five breed of sheep. Five breeds of male lambs used in the study were: Sumatera Composite breed (KS); Garut Composite breed sheep (KG); Barbados Cross breed sheep (BC); St Croix breed sheep (SC); and Local Garut breed sheep. The sheep fed on elephant grass ad libitum and supplemented by concentrate. Ten head of each breed were grouped into 4 groups according to body weight and assigned to one of diet treatments which was concentrate containing different levels of undegradable protein (UDP). The levels of UDP were 4.5% (UDP4.5) and 7.5% (UDP7.5). The UDP sources were from feed ingredients use to formulate concentrate, which was formulated in iso nitrogenous and iso energetic. Study was conducted in randomized complete block design in factorial 5 x 2 arrangement. Results shows that there was no interaction effect of breed and levels UDP and main effects of breed on N intake, N excretion, N absorption and N retention, ruminal pH, ammonia and blood urea N. UDP levels only significantly affected N intake and N urine. The N urine was significantly higher in UDP levels 4.5%. Similarly ruminal ammonia (NH<sub>3</sub>N) and blood urea nitrogen were significantly higher in UDP levels 4.5%. It can be concluded that five breeds of sheep had similar ability in N utilization of diet with different levels of UDP. Different levels of UDP produce similar positive N retention with average of 8.58 g/day.

**Keywords:** diet, nitrogen, sheep breeds, undegradable protein

## Introduction

Indonesian Research Institute for Animal Production developed composite breed of sheep which crossing between local breeds and exotic sheep breeds. The purpose of the crossing is to improve local breed performance. This composite breeds are: Barbados Black Belly Cross (BC); Garut Composite (KG) and Compass Agrinak (CA). The performance of these breeds affected by breed and environment. Yulistiani et al. (2014) reported that at village feeding management system and under stress condition, among four breeds of sheep, KG breed had better response to the better feed management. On the other hand CA breed, BC breed and local Garut sheep had lower growth rate and responded similarly to diet either unsupplemented or supplemented native grass basal diet. Similarly Subandriyo et al. (2000) also reported that CA breed and BC (as contemporary breed to Compass agrinak breed) had similar performance in trait of growth and dam productivity. The performance of these breeds can be increased when their nutrient requirement are sufficient.

Protein as one of critical nutrient required by ruminants, can be supplied from microbial protein synthesis and dietary protein that escapes from microbial degradation in the rumen (rumen undegradable protein/UDP) such as fish meal or blood meal reaches the small intestine for digestion and absorption. To support high rates of production, they must be supplied with UDP. However, the balance of UDP and RDP (rumen degradable protein) in the diet should be considered (Garg, 1998) so that able to support microbial growth which in turn increase fiber digestion in the rumen. McDonald et al (2002) suggested that for maintenance and growth of sheep, the RDP requirement was about 60% of total protein in the diet. So far, limited information on effect of different UDP content of diet on nitrogen utilization and ruminal fermentability of different breed of sheep. Therefore the objective of this study was to evaluate nitrogen utilization and ruminal fermentability of five breed of sheep fed on diet containing different levels of UDP.

## Materials and Methods

Study used 10 heads each from five breed of sheep, three breeds of which were from composite breeds of sheep developed in Balitnak namely: Barbados Black Belly Cross (BC) (genetic composition 50% Local

Sumatera 50% Barbados Black Belly), Garut Composite (KG) (50% Local Garut 25% St. Croix 25% Moulton Charolais), Compass Agrinak (CA) (50% Local Sumatera 25% St. Croix 25% Barbados Black Belly), and contemporary breeds were St. Croix (SC), and local sheep breed (Local Garut (LG)). The average body weight of sheep were  $21.01 \pm 3.24$ kg at age 11 months old. The animals in each breed were divided into two groups of diet treatments and were placed in individual pen during adaptation periods for diets treatment. The sheep were fed on fresh chopped elephant grass *ad libitum*. Supplements as diet treatment was offered at 450 g DM/day, were formulated to contain iso energetic (2.5 McalME/kg DM) and iso protein (16.7%) which contained two different levels of UDP (4.5 and 7.5%), and used as diet treatments. The feed ingredients used as UDP source in concentrate were coconut meal, rice bran, soybean meal, fish meal. Whereas part of RDP sources was from urea. The proportion UDP content of each feed was obtained from Mc Donald et al. (2002). Drinking water was freely available to the animals. During collection periods the sheep were moved to metabolic crates. Daily feed intake and refusal and fecal and urine output of the individual sheep were recorded. On the final day of collection period, ruminal fluid was collected from each of sheep 4 h after their morning feeding. Parameter recorded were N intake, N excretion (feces and urine), ruminal pH, ruminal ammonia, and plasma urea N. Study was conducted in completely randomized block design in 2 x 5 factorial arrangement, for two levels UDP (4.5 and 7.5%) and five breeds of sheep. Data were analyzed by ANOVA using General Linear Model procedure (GLM) of SAS 9.2 (2009). Differences among treatment means were detected using Duncan's multiple range tests.

## Results and Discussion

Nitrogen utilization of five breeds of sheep fed the different diets containing different levels of UDP are given in Table 1. There were no significant interaction between breeds and UDP levels on N intake, N excretion, N absorption and N retention. Therefore the data of main effect are presented in Table 1. Similarly, N utilization was also not significantly affected by breeds of sheep, N utilization among five breeds of sheep were similar. This indicated that these breeds have similar ability in utilizing N. Previously, Yulistiani et al (2015) reported that different breed of sheep had similar ability in digesting nutrients in the feed. Whereas UDP levels was only significantly ( $P < 0.05$ ) affected on N intake, N excretion in urine and percentage N excretion in urine. N intake was significantly higher in UDP 4.5%, however this higher intake was followed by higher N excretion in the urine, as results the percentage of urine excreted urine was also high in the concentrate contained UDP 4.5%. In this study in UDP 4.5% diet, part of degradable protein source is from urea while in diet UDP 7.5% contained higher true protein. Urea is rapidly degraded into  $\text{NH}_3\text{-N}$  (ammonia) in the rumen. When accumulation of ammonia exceed the optimum concentration needed by ruminal bacteria, some of the ammonia will be excreted in the urine. Therefore, in this study N excretion in urine was higher in diet UDP 4.5%. Though urine excretion was higher in UDP 4.5% but N retention was not significantly different between UDP 4.5 and 7.5% with average of 8.58 g/day.

Table 1. Nitrogen (N) utilization of five breed of sheeep fed on diet treatments

Factors	Parameter								
	N intake (g)	N excretion		N absorption (g)	N retention (g)	% of N intake			
		N Feses (g)	N urine (g)			N Feses	N urine	N absorption	N retention
Breeds									
Barbados Cross	18.30	4.48	4.93	13.82	8.88	24.38	26.87	75.51	48.75
St Croix	18.00	4.34	5.08	13.65	8.56	24.10	27.95	75.81	47.95
Compass Agrinak	18.37	5.19	5.00	13.18	8.18	28.22	27.05	71.71	44.72
Komposit Garut	18.46	4.91	4.87	13.55	8.67	26.45	26.22	73.41	47.31
Garut Local	18.14	3.91	5.60	14.23	8.62	21.35	30.70	78.44	47.94
UDP Levels									
4.5	18.72	4.46	5.64	14.23	8.57	23.78	30.05	76.04	46.16
7.5	17.79	4.65	4.54	13.14	8.60	26.00	25.47	73.91	48.50
SEM	0.4798	0.571	0.202	0.333	0.666	2.6636	3.0894	2.382	4.383
P values									
Breed	0.8703	0.223	0.806	0.107	0.8790	0.1363	0.6340	0.1362	0.907
UDP levels	0.0049	0.654	0.011	0.001	0.9720	0.1948	0.0259	0.1948	0.404
Breed x UDP levels	0.9292	0.646	0.936	0.759	0.7369	0.5900	0.9655	0.5900	0.741

There were no significant interaction between breed of sheep and UDP levels on ruminal pH, NH<sub>3</sub>-N concentration, and blood urea N taken at 0 and 4 hours after feeding, therefore the data are presented on main factors effect (Tabel 2). Ruminal pH taken at 0 and 4 hours after feeding were not affected by neither breed of sheep nor by UDP levels. Ruminal pH was similar across breeds and UDP levels. Ruminal NH<sub>3</sub>-N taken at 0 hour after feeding on the other hand was affected by breed of sheep, in which NH<sub>3</sub>-N of BC was significantly higher compared to GL. While, ruminal NH<sub>3</sub>-N taken at 4 hour after feeding was affected by UDP levels, in which the NH<sub>3</sub>-N concentration was significantly higher in sheep fed concentrate contained 4,5% UDP. Similar to ruminal NH<sub>3</sub>-N taken at 4 hour, BUN was also affected by UDP levels. The BUN of UDP 4.5% was significantly higher than UDP 7.5%. Ruminal ammonia concentration is related to degradability of protein in the rumen. As previously mention that source of degradable protein in diet UDP 4.5% partly is from urea which is rapidly degraded in the rumen and converted into ammonia. Therefore the ruminal ammonia concentration particularly at 4 hours after feeding in UDP 4.5% was significantly higher than UDP 7.5%. The concentration of ruminal ammonia of both diet was in the range of optimum concentration (8.5 – 30 mg/dl) for rumen microbial activity (McDonald et al 2002). Similar result was also reported by Kang et al (2015) that urea supplementation as protein source in rice straw basal diet produced higher ruminal ammonia than soybean meal supplementation. Blood urea nitrogen is derived from absorbed ruminal NH<sub>3</sub>-N, therefore the increase ruminal ammonia also resulted in increasing levels of blood urea nitrogen. Concentrations of BUN are highly positively correlated to the level of NH<sub>3</sub>-N production in the rumen therefore sheep fed higher ruminal degradable protein (UDP 4.5% ) in this study had higher BUN than in UDP 7.5%.

Table 2. Rumen pH, NH<sub>3</sub>-N concentration (mg/dl), and blood urea N taken at 0 and 4 hours after feeding of five breed of sheep fed on diet treatments

Factors	0 hours		4 hours		Blood urea nitrogen
	pH	NH <sub>3</sub> -N	pH	NH <sub>3</sub> -N	
Breeds					
Barbados Cross	6.64	11.12	6.12	19.64	55.85
St Croix	6.81	9.09	6.20	16.07	45.87
Compass Agrinak	6.79	9.81	6.25	18.65	53.75
Komposit Garut	6.80	9.32	6.21	17.87	52.62
Garut Local	6.79	7.7	6.10	17.76	49.12
UDP levels					
4.5	6.82	9.86	6.23	20.49	55.0
7.5	6.71	8.96	6.13	15.51	47.89
SEM	0.0988	0.6010	1.1297	1.7023	2.5322
P values					
Breed	0.3989	0.032	0.748	0.7932	0.3689
UDP levels	0.1082	0.1723	0.2168	0.0108	0.0001
Breed x UDP levels	0.8227	0.6879	0.9550	0.9292	0.9292

## Conclusion

Different breed of sheep use in this study had similar ability in utilizing nitrogen in the diet containing different levels of UDP. In iso energetic and iso protein diet different level of UDP content, had similar nitrogen utilization but lower UDP content (UDP 4.5%) produced higher rumen ammonia and blood urea nitrogen due to urea in the diet.

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# A Willingness to Pay Evaluation of Silage Implementation for Small Dairy Farmers in Central & East Java

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## Abstract

*The objectives of the field research were to examine the silage adoption level by small dairy farmers located in Malang, Boyolali, and Wonosobo districts, its challenge and opportunities. One of underlying causes of low milk production (on average 10-15 liter per cow per day) by small farmers in Indonesia is the lack of forage mainly during dry season. This problem root worsening by the increasingly narrower owned land convert to non-agricultural activities. Silage is a simple technology to conserve the surplus of grass during the rainy season in order to able to feed good quality grass during the dry season. There are 60 small dairy farmers as respondents from 3 districts were deeply interviewed about their forage supply along the year, what they do if they are lacking of forages, their knowledge about silage, their experience on producing and feeding silage and how much price they are willing to pay if a vendor produce silage commercially. The information were analyzed using descriptive statistic in SPSS program software version 22 was used to analyzed the data exported from Akvo dashboard. To implement the silage feeding on small farmers faced some challenges including lack of skill, not all farmers having grass surplus, not all farmers having enough capital to purchase chopper, and lack of land to store the silage. Those challenges is main reason why the willingness to pay survey was done. The results show in Boyolali district, there are 67% farmers who are lacking of forages are willing to buy silage, while in Malang district, about 57% are willing to buy. In contrast, in Wonosobo district, the willingness to buy silage is higher from farmers who have sufficient forages where all respondents (100%) stated willing to buy silage. While only about 72.7% farmers from inadequate forage supply stated that, they want to buy silage. More than 50% respondents are willing to pay silage, meaning as the opportunity to dairy cooperative, or company or farmer group to produce and sell it to small farmers as a new business unit.*

*Keywords: forage, silage, small farmers*

## Introduction

About 97% dairy population are located in Java Island. The highest population is in East Java (45.6%), followed by Central Java (27.7%) and East Java (23.5%) (Politeknik Jember 2015) with the total population is about 480,000 cows in 2014 growing 21.7% from 2013 (BPS 2015) so that it is very important to improve the dairy farming practice in Java in order to increase milk production.

The main problem of milk production in Indonesia is on average low milk production per cow (about 11 liters per cow per day), small number of cows kept in each farmer, limited grass land, not yet fully applied good farming practice and lack of supervision (Boediyono 2008). The main cause of low milk production is related to feeding practice whether cows as ruminant animal are able to obtain sufficient forages or not.

Silage, is an alternative conserved forage can be made from grass, maize plant, and sugarcane leaves. Silage technology originally from temperate zone when during winter there is no grass available. In Indonesia, silage has been introduced more than two decades as alternative good quality forage during dry season. However the application of silage feeding by dairy farmers are very rare mainly from eastern Indonesia where actually the farmers there need silage more because of the longer dry season (Umiyasih & Wina 2008).

Permana & Tanjung (2013) during dairy pilot project West and East Java in SNV found that the silage implementation on small dairy farmers having some obstacles as follow: many farmers do not have grass surplus, do not have choppers and labor to produce silage. The field research is done to investigate how much the price that dairy farmers willing to pay if silage is sold to the market by dairy cooperative or feed company.

## Materials and Methods

The field research using depth interviews with 55 dairy farmers was conducted in three districts namely Boyolali, Malang and Wonosobo. Taking respondents using cluster random sampling. In Boyolali,

Respondents are from 4 clusters from 3 dairy cooperatives covering area, namely KUD Boyolali, KUD Mojosoongo, KUD Cepogo, and Selo Subdistrict. Same technique using in Wonosobo, two clusters from Kapencar and Purbosono villages in Kertek and three clusters from Bumitirto, Semayu and Plobangan villages in Selomerto subdistrict. In total, 20 respondents were interviewed from these subdistricts. In Wonosobo district there has no dairy cooperative established yet. In Malang, the respondents are from KAN Jabung dairy farmers in Jabung subdistricts.

Data was collected using device and mobile survey-AKVO application and by surveyor. Collected data was analysed using descriptive statistic in SPSS program software version 22 was used to analysed the data exported from Akvo dashboard. The data were cleaned before performing analysis using the distribution of frequency of all variables to identify missing and in consistency.

## Result and Discussion

In line with other previous studies, the lack of forage is the main problem in small dairy farmers. Here 80% respondents stated that they are lack of forages mainly during the dry season, while only 11% is sufficient. Possibly those are farmers who have access to rent land or owned bigger size of grass land.

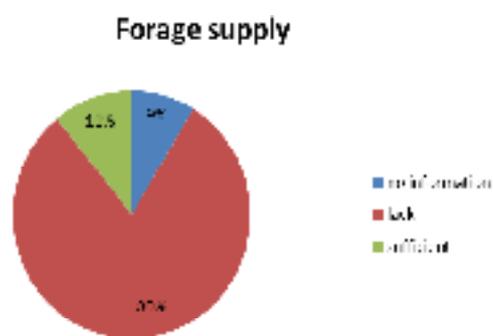


Figure 1. Forage supply in small dairy farmer

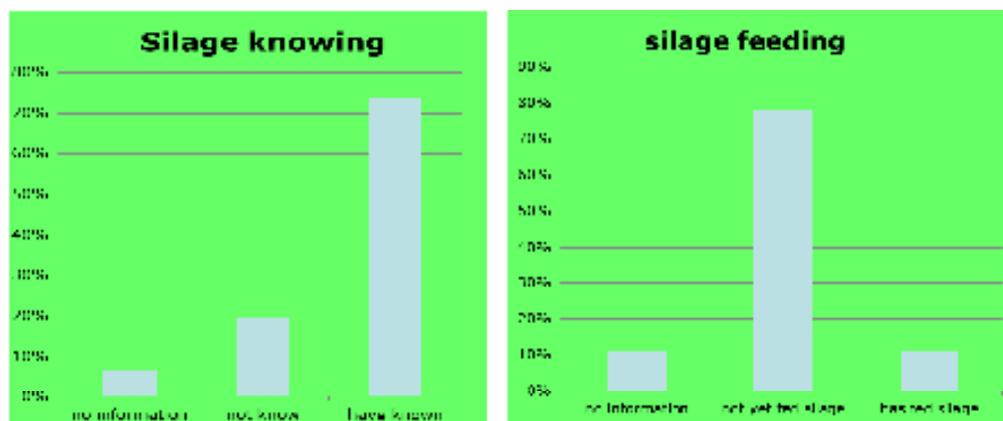


Figure 2 and 3. Silage knowing and feeding by small dairy farmers (Baseline Malang 2014)

Since silage research and introduction have been done quite long ago (Erowati 2000), Wina (2005). Majority farmers have known about silage. However almost all respondent have not fed silage to their cows due to several causes including not all farmers having equipment for silage production, having surplus grass, and limited land to store silage.

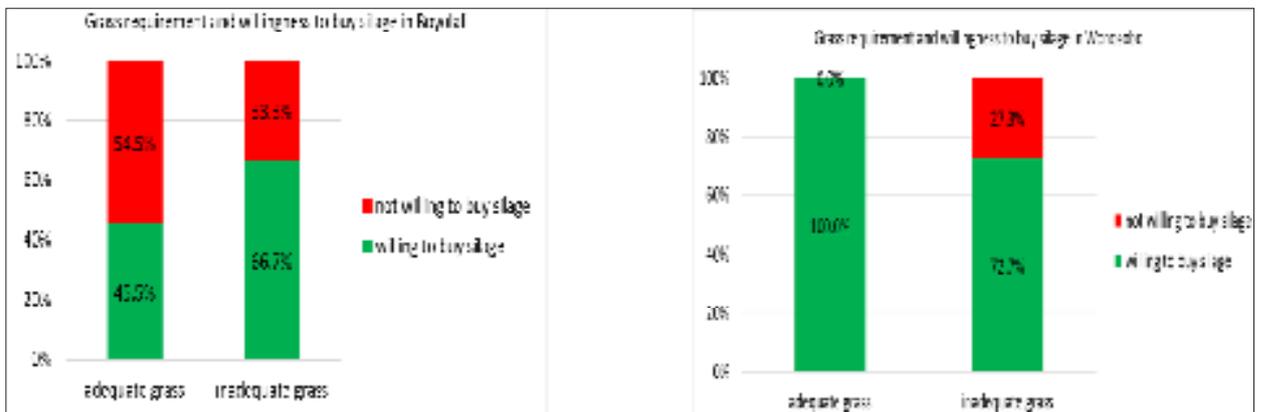


Figure 4 and 5. Grass sufficiency and willingness to pay for silage by small dairy farmers

From figures above, seen that dairy farmers are potential buyer showing the demand if there is a company or dairy cooperative produce silage mainly during the dry season. Similar result for Malang district, where about 57% respondents said that they want to buy silage. The pilot project in 2014 that conducted by SNV in collaboration with NESTLE, PIONEER and KAN Jabung showed that the silage sales reached the peak during the dry season as described in the Figure 6.

Silage provided by dairy cooperatives or company in commercially is one method to increase silage adoption. Silage technology adoption by small dairy farmers to cope lack of good quality forages mainly during the dry season. By implementing silage can preserve the forages quality.

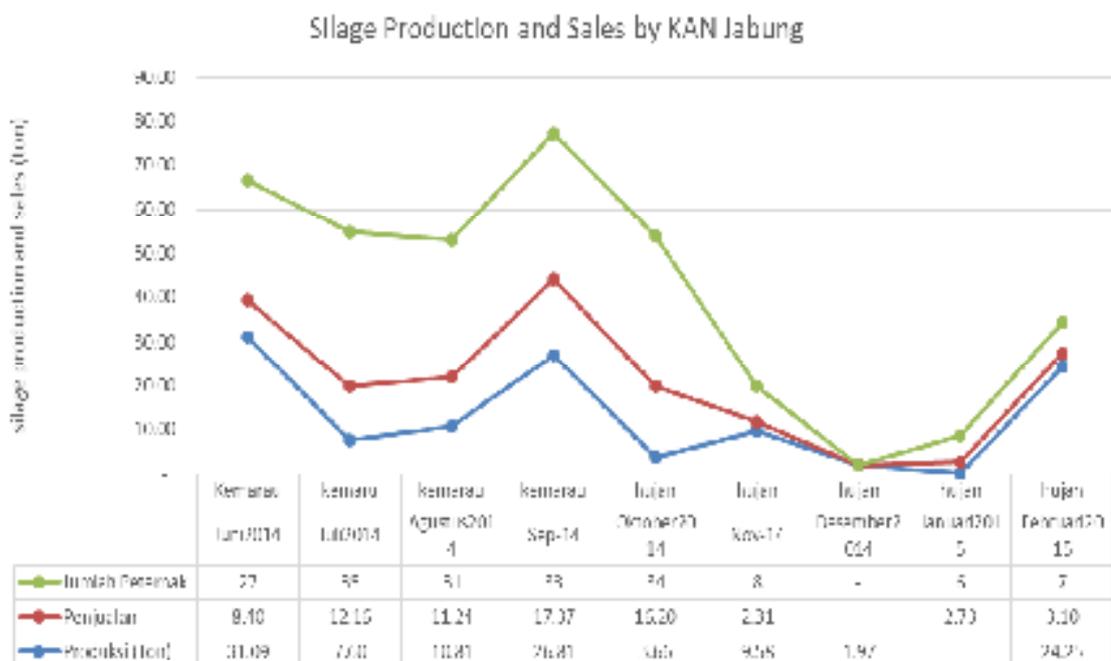


Figure 6. Number of farmers, silage production and sales in KAN Jabung, Malang in 2014

## Conclusion

To produce silage commercially by dairy cooperatives may increase the silage adoption by small dairy farmers. The benefits are standardized silage quality, more economically feasible because producing at larger scale, easy access for dairy farmers to buy with refill system using plastic drum.

## Acknowledgment

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# Fermentability and Digestibility of Rice Straw-Concentrate Base Ration Added with Probiotic

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## Abstract

Rice straw - concentrate base ration has low fermentability and digestibility that could not support animal performance, especially beef cattle. In fact, such kind of ration is usually given by farmers. Adding commercial solid or liquid probiotics into that ration can improve its utilisation resulting positive effects on beef cattle performance. Effect of solid and liquid probiotic addition on rumen fermentation and digestibility has also been studied, but the effect was influenced by level and type of probiotic addition. Probiotic level was 0.25% w.w<sup>-1</sup> for solid, and 0.10% v.w<sup>-1</sup> for liquid probiotics. In this experiment, probiotic levels were increased and its effects on *in vitro* fermentability and digestibility of rice straw - concentrate base ration were studied. A randomised block design was used; treatments were A1 (rice straw - concentrate base ration as control treatment), A2 (A1 + 1.25% w.w<sup>-1</sup> solid probiotic) and A3 (A1 + 0.5% v.w<sup>-1</sup> liquid probiotic); rumen fluid from five beef cattles was used as replications. Analysis of variance and orthogonal contrast were applied. Ammonia and VFA concentrations from all treatments were similar at 0 h incubation. Probiotic addition did not influence ammonia and VFA concentrations at 2 h incubation. Treatments affected DM and OM digestibilities, but no significant differences between liquid and solid probiotic effects. Therefore, adding probiotic improved digestibility of rice straw - concentrate ration; however, improvement in rumen fermentation could occur after 2 h incubation.

*Keywords:* concentrate, digestibility, fermentability, probiotic, rice straw

## Introduction

Rice straw - concentrate base ration has low fermentability and digestibility that could not support animal performance, especially beef cattle. In fact, such kind of ration is usually given by farmers. Adding commercial solid (0.25% w.w<sup>-1</sup>) or liquid (0.10% v.w<sup>-1</sup>) probiotics (on concentrate basis) into that ration can improve its utilisation resulting positive effects on beef cattle performance (Suryahadi *et al.*, 2012). These occurred through its effects on balancing the microbial populations, increasing microbial activity and producing active substances that improve condition in digestive tract for nutrient fermentation and digestion (Dawson, 1993; Kamra and Pathak, 1996; Suroño, 2004). Effect of solid and liquid probiotic addition on rumen fermentation and digestibility of rice straw or elephant grass - concentrate rations has also been studied (Kristina, 2013; Septiani, 2013; Almai, 2013), but the effect was influenced by level and type of probiotic addition; liquid probiotic could be used up to 0.20% v.w<sup>-1</sup>, and solid probiotic up to 0.50% w.w<sup>-1</sup> (Muzakki, 2014; Rafi, 2014). In this experiment, probiotic levels were increased and its effects on *in vitro* fermentability and digestibility of rice straw - concentrate base ration were studied.

## Materials and Methods

*In vitro* fermentation and digestibility experiments were conducted following Tilley and Terry (1963) procedure modified by Sutardi (1979). Treatments were A1 (rice straw - concentrate base ration as control treatment), A2 (A1 + 1.25% w.w<sup>-1</sup> solid probiotic) and A3 (A1 + 0.50% v.w<sup>-1</sup> liquid probiotic). Solid (Biofeed) and liquid (Turrimavita) probiotics (commercial products produced by CV Sinar Aras) used in this experiment were the same as those were used by Suryahadi *et al.* (2012). Each treatment ration (1 g) was mixed with 12 mL artificial saliva (McDougall, 1948) and 8 mL rumen fluid in a fermentation tube closed with a rubber stopper. The mixture was fermented anaerobically after gassing with CO<sub>2</sub> at 39 °C (pH 6.8) in a shaker water bath. Fermentation was stopped after 0 and 2 h by adding saturated HgCl<sub>2</sub> solution (2 drops), fermentors were centrifuged (3000 rpm; 15 min). Supernatants were used to determine ammonia and VFA concentrations, respectively following Conway microdiffusion and steam distillation methods (Department

of Dairy Science, University of Wisconsin, 1966). The same procedure was applied for digestibility experiment, but the mixtures were fermented anaerobically for 24 h. After stopping fermentation by using saturated HgCl<sub>2</sub> solution, and centrifuging (3000 rpm; 15 min), the residues were added with pepsin-HCl solution (20 mL) and incubated aerobically (39 °C; 24h). Each mixtures were then filtered through a Whatman filter paper No. 41 using a vacuum pump. Ration samples and residues were dried in an oven (105 °C; 24 h) to determine its dry matter (DM) content, then were ashed in a furnace (600 °C; 6 h) to measure ash content and calculate organic matter (OM) content.

A randomised block design was used and rumen fluid from five beef cattles was used as replications. Analysis of variance and orthogonal contrast test were applied (Steel and Torrie, 1993). Variables measured were ammonia and total VFA concentrations at 0 and 2 h incubations, and DM and OM digestibilities.

## Results and Discussion

Solid probiotic had total plate count (TPC)  $3.9 \times 10^8$  colony forming unit (CFU).gram<sup>-1</sup>, and composed of *Lactobacillus* sp. ( $7.2 \times 10^9$  CFU.gram<sup>-1</sup>), *Bifodobacterium* sp. ( $4.9 \times 10^9$  CFU.gram<sup>-1</sup>), *Streptococcus* sp. ( $5.6 \times 10^7$  CFU.gram<sup>-1</sup>) and *Bacillus* sp. ( $4.0 \times 10^5$  CFU.gram<sup>-1</sup>). Liquid probiotic contained  $1.5 \times 10^{10}$  CFU.gram<sup>-1</sup> with populations of *Lactobacillus* sp. :  $1.1 \times 10^{10}$  CFU.mL<sup>-1</sup>, *Bifodobacterium* sp. :  $7.0 \times 10^5$  CFU.mL<sup>-1</sup> and *Streptococcus* sp. :  $1.0 \times 10^{10}$  CFU.mL<sup>-1</sup>; however, *Bacillus* sp. was detected in very small numbers in liquid probiotic (Suryahadi *et al.*, 2012). These populations and bacterial species compositions had met the requirement for its used as probiotic,  $10^6$  cfu.g<sup>-1</sup> (Tamime *et al.*, 2005). Using 0.25% w.w<sup>-1</sup> solid and 0.10% v.w<sup>-1</sup> liquid probiotics alone did not cause differences on *in vitro* fermentation (Kristina 2013).

Control ration composed of rice straw and concentrate at a ratio of 60 : 40% DM basis. Calculated nutrient composition indicated that control ration contained 60.24% DM, 20.93% ash, 8.49 % crude protein, 3.35% ether extract, 10.44% crude fibre, 56.79% non nitrogen free extract, 61.46% TDN, 0.80% Ca and 0.29% P (on DM basis). Crude protein and energy contents of control ration were still below the energy (70% TDN) and crude protein (12 - 13%) contents suggested by NRC (2000), but other nutrient contents had met the nutrient requirement for beef cattle (430 kg body weight).

Ammonia and VFA concentrations from all treatments were similar at 0 h incubation; ammonia and VFA concentrations in all treatments increased after 2 h incubation (Table 1). However, probiotic addition did not influence ammonia and VFA concentrations at 2 h incubation. Treatments affected DM and OM digestibilities which were increased by probiotic supplementation; however, there was no significant difference between solid and liquid probiotics on DM and OM digestibilities.

Table 1. Effect of probiotic supplementation on *in vitro* fermentability and digestibility of rice straw concentrate base diet

Variables	Control ration	Control ration + 1.25% solid probiotic	Control ration + 0.50% liquid probiotic
Ammonia concentration (mM)			
0 h	3.65 ± 0.64	3.96 ± 0.58	3.99 ± 1.68
2 h	6.24 ± 0.49	6.33 ± 1.23	6.95 ± 2.72
VFA concentration (mM)			
0 h	52.15 ± 5.81	53.10 ± 5.19	58.79 ± 12.36
2 h	65.43 ± 13.16	79.65 ± 5.19	80.60 ± 13.41
DM digestibility (%)	35.37 ± 5.13 <sup>a</sup>	38.27 ± 4.89 <sup>b</sup>	41.43 ± 4.51 <sup>b</sup>
OM digestibility (%)	42.24 ± 6.14 <sup>a</sup>	44.40 ± 5.75 <sup>b</sup>	48.15 ± 5.33 <sup>b</sup>

Means with different superscript at the same row differ at P<0.05 on the basis of orthogonal contrast test

At the beginning (0 h incubation), the starting concentrations for ammonia and VFA were similar in all treatments and the effects of probiotic addition had not yet occurred. Fermentation by rumen microbes occurred at 2 h incubation indicated by increases in ammonia and VFA concentrations. However, adding probiotic did not increase protein degradation and energy source fermentation of rice straw - concentrate base diet. This differs from that was obtained by Kristina (2013), ammonia concentrations (5.20 mM) of similar control ration and that was added with solid probiotic (0.25% w.w<sup>-1</sup>) were lower than that (6.12 mM) of control ration added with liquid probiotic (0.10% v.w<sup>-1</sup>). Adding liquid probiotic also increased VFA concentration (149.35 mM) with VFA concentrations for control ration and that added with solid probiotic

were respectively 107.10 and 114.51 mM. Effect of liquid probiotic in affecting rumen fermentation occurred through stimulation of more population of rumen bacteria, but no significant effect on protozoal population in comparison to solid probiotic (Kristina, 2013). This means that bacterial species in liquid probiotic were more adaptable to rumen condition and capable of stimulating growth and enzyme activity of rumen bacteria than those in solid probiotic, consequently, increasing protein degradation and energy source fermentation. However, that situation did not occur in this experiment which may probably relate to the batch of probiotics, differences in sample and nutrient content of rice straw - concentrate ration, and differences in experimental condition (France dan Dijkstra 2005; Muzakki, 2014).

Except for ammonia concentrations at 0 h incubation, ammonia concentrations of all treatments were in the range of those suggested by Sutardi (1979), 4 - 12 mM, and McDonald *et al.* (2002), 6 - 21 mM, meaning that ammonia concentration was still sufficient for rumen microbial protein synthesis. In contrast, VFA concentrations at 0 and 2 h were lower than or in the minimum range of the normal level suggested by McDonald *et al.* (2002), 70 - 150 mM. This demonstrates that the ratio between ammonia and VFA concentrations may not in optimum ratio for microbial protein synthesis in the rumen.

In this experiment, solid and liquid probiotic addition increased DM and OM digestibilities at a similar extent. This shows that both probiotics may affect bacterial and other rumen microbial growth and population, as well as its enzyme activity in rumen fermentation of rice straw - concentrate ration in longer incubation period (more than 2 h). This result was similar to that was obtained by Kristina (2013); probiotic use did not increase rate of protein degradation and energy source fermentation within 2 h incubation. Result of Muzakki (2014) indicates that ammonia and VFA concentrations of similar control ration added with liquid probiotic (0, 0.1 and 0.2% v.w<sup>-1</sup>) increased linearly from 0, 1, 2 and 3 h incubation. However, the same results were not obtained when solid probiotic (0, 0.25 and 0.50% w.w<sup>-1</sup>) was used in similar control ration incubated for 0, 1, 2, and 3 h (Rafi, 2014). In addition, fermentation of rice straw needs longer time due to its crude fibre, lignin and silica contents making difficulties in nutrient fermentation (Sutardi, 1980; Rinduwati and Ismartoyo, 2002; Sarnklong *et al.*, 2010). These situations demonstrate that significant effect of probiotics in affecting protein degradation and energy source fermentation of rice straw - concentrate diet may occur after 3 h and within 24 h. This is because the fermentation products of that ration added with probiotic after 24 h incubation could be digested by pepsin enzyme increasing DM and OM digestibilities. Increases in DM and OM digestibilities of rice straw - concentrate ration adding with liquid probiotic were also obtained by Kristina (2013) and Muzakki (2014), but it could not be produced when using solid probiotic (Rafi, 2014). Although increases in DM and OM digestibilities varied among experiments, this experiment was still capable of showing positive effects of adding solid (1.25% w.w<sup>-1</sup>) and liquid (0.5% v.w<sup>-1</sup>) probiotics in improving DM and OM digestibilities of control diet.

## Conclusion

Probiotic supplementation improved DM and OM digestibilities of rice straw - concentrate ration; however, improvement in rumen fermentation could occur after 2 h incubation.

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# Effects of Solid or Liquid Probiotic Supplementation on Rumen Microbial Population and Enzyme Activity

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## Abstract

Probiotics are live microbial feed additive which beneficially affect the host by improving its intestinal microbial balance. Solid and liquid probiotics have been used as feed additive for beef cattle and its roles had been studied *in vivo* and *in vitro*. However, there is still limited study about effects of probiotic utilisation on rumen microbial populations and enzyme activity that improve feed fermentation and utilisation in the rumen. The aim of the experiment is to study effect of solid and liquid probiotic supplementation on rumen protozoal and bacterial populations, and its enzyme activity. An *in vitro* experiment was conducted using randomized block design with five replications. The treatments consisted of A0 (rumen fluid without ration and probiotic supplementation as negative control), A1 (control ration without probiotic supplementation as positive control), A2 (A1 + solid probiotic supplementation at 1.25% w.w<sup>-1</sup>) and A3 (A1 + liquid probiotic supplementation at 0.5% v.w<sup>-1</sup>). Variables were populations of protozoa and bacteria (total bacteria, amylolytic, proteolytic, cellulolytic, lactic acid, and lipolytic bacteria), and enzymes activity (amylase, cellulose, protease, and lipase activity). Use of liquid probiotic increased amylolytic and lipolytic bacteria at 2 h incubation, but did not affect populations of protozoa, total bacteria, cellulolytic, proteolytic and lactic acid bacteria. No significant effects of treatments on all enzyme (amylase, cellulose, protease, and lipase) activity, activities. It is concluded that probiotic supplementation could only affect amylolytic and lipolytic bacteria and the use of liquid probiotic was more effective than solid probiotic. We can suggest that supplementation of liquid probiotics was more effective to increase amylolytic and lipolytic bacteria.

**Keywords:** bacteria, enzyme, liquid probiotic, protozoa, solid probiotic

## Introduction

Probiotics are live microbial feed additive which beneficially affect the host by improving its intestinal microbial balance (Fuller, 1992). Solid and liquid probiotics have been used as feed additive for beef cattle; its roles had been studied *in vivo* and *in vitro*. Given solid and liquid probiotics *in vivo* did not affect nutrient intakes of rice straw - concentrate base diet, except for ether extract (EE) intake (Purwanti, 2013); improved its dry matter (DM), crude protein (CP) and crude fibre (CF) digestibilities with liquid produced better results than solid probiotics (Siregar, 2013), and reduced faecal ammonia concentration (Muchayani, 2013). Liquid probiotic had better effects in increasing *in vitro* total bacterial population, ammonia and VFA concentrations, and DM and OM digestibilities of rice straw - concentrate base diet than that of solid probiotic (Kristina, 2013; Almai, 2013); in contrast, Septiani (2013) showed that solid probiotic had better effects on fermentation and digestibility of elephant grass - concentrate base diet. Probiotic effect was also influenced by addition level which were up to 0.20% v.w<sup>-1</sup> and 0.50% w.w<sup>-1</sup>, each for liquid and solid probiotics (Muzakki, 2014; Rafi, 2014). Probiotic uses had been studied focusing on fermentation and digestibility; however, there is still limited study about effects of probiotic utilisation on rumen microbial populations and enzyme activity that can improve feed fermentation and utilisation in the rumen. Therefore, this experiment is aimed at studying effect of solid or liquid probiotic supplementation on rumen protozoal and bacterial populations, and its enzyme activity.

## Materials and Methods

*In vitro* fermentation experiment was done using the first of two stages of Tilley and Terry method (1963) modified by Sutardi (1979). Fermentation was carried out in a fermentor tube containing each of treatment diet (1 g), McDougall (1948) artificial saliva solution (12 ml) and rumen fluid (8 ml), closed with a rubber stopper after gassing with CO<sub>2</sub>. Fermentation was done anaerobically (39 °C, pH 6.8) for 0 and 2 h in a shaker water bath. Samples were taken at 0 and 2 h for counting bacterial and protozoal populations.

Fermentation was then stopped by adding saturated HgCl<sub>2</sub> solution (2 drops). The tubes were centrifuged (3000 rpm, 15 min); supernatants were used for analysis of enzyme activity.

Bacterial populations were counted after serial dilution of each sample (Ogimoto and Imai, 1981). Diluted sample was cultured onto brain heart infusion (BHI) solid medium for total bacterial population, BHI solid medium added with each substrate (starch, carboxymethyl cellulose, olive oil and skim milk for amylolytic, cellulolytic, lipolytic and proteolytic bacteria), and de-Mann Rogossa Sharpe (MRS) solid medium for lactic acid bacteria. Analysis of cellulase and amylase activities were done with Tripathi and Karim (2011) method. Lipase activity was determined with titrimetric method (Nurhasanah and Herasari, 2008), and protease activity was analysed with Bergmeyer and Gawhn (1983) method described by Kurniawati (2007).

A randomised block design was applied (five replications of beef cattle rumen fluid). Treatments: A0 (rumen fluid without diet and probiotic supplementation=blank), A1 (control diet without probiotic supplementation=positive control), A2 (A1+ 1.25% w.w<sup>-1</sup> solid probiotic) and A3 (A1+ 0.5% v.w<sup>-1</sup> liquid probiotic). Commercial probiotic products from CV Sinar Aras were used (Biofeed=solid probiotic; Turrimavita=liquid probiotic). Variables: protozoal and bacterial (total bacteria, amylolytic, cellulolytic, lipolytic, proteolytic and lactic acid bacteria) populations, and enzyme activities (amylase, cellulase, lipase and protease activities). Data were analysed with analysis of variance and contrast orthogonal (Steel and Torrie, 1993).

## Results and Discussion

Total plate count and each of probiotic bacterial populations (*Lactobacillus* sp., *Bifidobacterium* sp., *Streptococcus* sp. and *Bacillus* sp.) in solid and liquid probiotics can be seen in Suryahadi *et al.* (2012). Calculated nutrient composition of control diet (rice straw and concentrate 60 : 40% dry matter, DM, basis) was 60.24% DM, 20.93% ash, 8.49 % CP, 3.35% EE, 10.44% CF, 56.79% non nitrogen free extract, 61.46% TDN, 0.80% Ca and 0.29% P. The protein and energy contents were lower than NRC (2000) recommendation for beef cattle weighing at 430 kg.

Microbial populations at 0 h were similar in all treatments, except for cellulolytic bacteria (Table 1); fermentation of treatment diets occurred at similar microbial populations and condition. Differences in fermentation among treatments were expected due to treatment effects. Differences in cellulolytic bacteria at 0 h may indicate cellulolytic bacteria were the main bacteria remained in rumen fluid of beef cattle before the cattles were slaughtered, and addition of rice straw - concentrate diet may stimulate those bacteria. However, populations of those bacteria in all treatments were not change significantly after 2 h incubation meaning that probiotic effects on cellulolytic bacterial population may occur in longer period than 2 h. This could be similar to of probiotic effect on cellulase activity (Paul *et al.*, 2004).

Table 1. Populations of protozoa and bacteria

Populations (log cfu.ml <sup>-1</sup> rumen fluid)	Incubation period (h)	Treatments			
		Blank	Control diet	Control diet + solid probiotic	Control diet + liquid probiotic
Protozoa (log sel.ml <sup>-1</sup> )	0	3.93 ± 0.29	4.05 ± 0.49	4.15 ± 0.57	4.09 ± 0.52
	2	3.70 ± 0.68	3.83 ± 0.62	3.76 ± 0.50	3.82 ± 0.54
Bacteria :					
Total	0	6.05 ± 0.72	6.65 ± 0.32	6.73 ± 0.39	6.75 ± 0.52
	2	6.44 ± 0.32 <sup>b</sup>	6.94 ± 0.17 <sup>a</sup>	6.58 ± 0.70 <sup>b</sup>	6.81 ± 0.23 <sup>b</sup>
Amylolytic	0	6.44 ± 0.20	6.83 ± 0.72	7.26 ± 0.93	7.44 ± 1.20
	2	6.37 ± 0.60 <sup>c</sup>	7.80 ± 0.47 <sup>a</sup>	6.82 ± 0.90 <sup>c</sup>	7.27 ± 0.83 <sup>b</sup>
Cellulolytic	0	6.24 ± 0.38 <sup>c</sup>	6.61 ± 0.28 <sup>b</sup>	6.73 ± 0.39 <sup>a</sup>	6.71 ± 0.47 <sup>a</sup>
	2	6.24 ± 0.80	6.96 ± 0.18	6.68 ± 0.58	6.82 ± 0.25
Lipolytic	0	6.36 ± 0.49	6.44 ± 0.35	6.65 ± 0.50	6.64 ± 0.49
	2	6.09 ± 0.77 <sup>b</sup>	6.77 ± 0.39 <sup>a</sup>	6.50 ± 0.85 <sup>b</sup>	6.86 ± 0.24 <sup>a</sup>
Proteolytic	0	6.46 ± 0.33	6.63 ± 0.18	6.76 ± 0.35	6.61 ± 0.50
	2	6.11 ± 0.80 <sup>b</sup>	6.86 ± 0.20 <sup>a</sup>	6.48 ± 0.77 <sup>b</sup>	6.70 ± 0.32 <sup>b</sup>
Lactic acid	0	6.04 ± 0.65	6.13 ± 0.49	6.36 ± 0.62	6.41 ± 0.71
	2	5.92 ± 0.29 <sup>b</sup>	6.81 ± 0.21 <sup>a</sup>	± 0.75 <sup>b</sup>	6.29 ± 0.84 <sup>b</sup>

Means with different superscript differ at P<0.05 based on contrast orthogonal test

Protozoal populations were not affected by probiotic addition at 2 h incubation (Table 1) which was similar to that obtained by Kristina (2013). No differences in protozoal populations at 0, 2, 4, 6 and 8 h were found by Abe and Iriki (1977); its populations in this experiment were still in the normal range,  $10^4$  -  $10^6$  (Koike and Kobayashi (2009). Table 1 also shows that bacterial populations differed among treatments at 2 h incubation, except for cellulolytic bacteria. The greatest populations of all bacteria were found in control diet with the lowest were observed in blank sample. Probiotic addition did not significantly increase total, proteolytic and lactic acid bacterial populations compared to blank. Liquid probiotic stimulated greater populations of amylolytic and lipolytic bacteria to similar populations to those in control diet than solid probiotic. Probiotic bacteria in liquid probiotic were more adaptable to rumen condition that stimulate growth of amylolytic and lipolytic bacteria (having faster growth rate than the other bacteria) in rumen fluid containing rice straw - concentrate base diet. The same results were obtained by the others (Kristina, 2013; Almai, 2013; Siregar, 2013).

Enzyme activities were not different among treatments at 0 h incubation, except for lipase activity with blank had the lowest lipase activity (Table 2). After 2 h incubation, enzyme activity in all treatments remained the same. This means that alterations in all of enzyme activities that we examine may require longer incubation time. The addition of probiotics were not increase enzyme activities. Effects of probiotic additions could occur after 2 h incubation. Present results were in the same agreements as those found by Dhital *et al.* (2014) showing that a complete starch granule fermentation occurred in more than 24 h incubation; Paul *et al.* (2004) indicating that probiotic addition might stimulate cellulase activity after 24 - 48 h incubation; and Selje-Assmann *et al.* (2007) demonstrating that proteolytic activity in degrading casein protein was optimum at 4 h incubation. Differences in lipase activity between blank and the others could be due to the addition of concentrate in treatment diet that may stimulate lipase activity from the rumen microbes (Latham *et al.*, 1972).

Table 2. Enzyme activity

Enzyme activity (unit.g <sup>-1</sup> DM)	Incubation period (h)	Treatments			
		Blank	Control diet	Control diet + solid probiotic	Control diet + liquid probiotic
Amilase ( $10^{-6}$ )	0	1.26 ± 0.87	1.02 ± 0.48	1.10 ± 0.43	1.10 ± 0.32
	2	1.47 ± 0.91	1.08 ± 0.39	1.03 ± 0.32	1.17 ± 0.49
Cellulase ( $10^{-6}$ )	0	1.17 ± 1.02	1.16 ± 0.74	1.06 ± 0.59	1.22 ± 0.66
	2	1.40 ± 1.10	1.35 ± 0.96	1.01 ± 0.44	1.31 ± 0.81
Lipase	0	0.09 ± 0.04 <sup>b</sup>	0.14 ± 0.07 <sup>a</sup>	0.14 ± 0.54 <sup>a</sup>	0.14 ± 0.06 <sup>a</sup>
	2	0.11 ± 0.03	0.14 ± 0.07	0.14 ± 0.06	0.14 ± 0.05
Protease	0	4.94 ± 4.17	5.64 ± 3.67	4.68 ± 3.07	6.04 ± 4.98
	2	3.95 ± 3.52	4.73 ± 3.56	4.22 ± 4.32	3.69 ± 3.79

Means with different superscript differ at  $P < 0.05$  on the basis of contrast orthogonal

## Conclusion

Probiotic supplementation could only affect amylolytic and lypolytic bacteria and the use of liquid probiotic was more effective than solid probiotic. Addition of 1.25% w.w<sup>-1</sup> solid or 0.5% v.w<sup>-1</sup> liquid probiotics have not been sufficient to increase all enzymes (amylase, cellulose, protease, and lipase) activities at 2 h incubation. The positive effects of probiotic addition on microbial populations and its enzymes activities could occur in more than 2 h incubation period.

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# Effect of Ammoniated Straw on Methane Production in an *in vitro* System and on Growth Performance

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## Abstract

Egyptian desert is considered as a source of fodder along the year. These plants had a lot of nutritive problems need to solve before depending on it as a good quality fodder. Objectives: to evaluate the effects of using different combination of tannin ferrous plants with ammoniated wheat straw at different ratio on methane production and lamb performance. Methodology: An *in vitro* incubation system was used. Results: most tested blood parameters and Barki lambs daily body gains were significantly affected by tested rations. Consumption of combination of tanniniferous plant containing varied classes of secondary compounds (e.g., tannins and spooning) with resource of energy as cassava lead to reduce their toxic effects. Conclusion: improving diet quality can both improve animal performance and efficiency by reducing methane emissions per unit of animal product.

Keywords: ammoniated straw, local Barki sheep, methane production, tannin plants

## Introduction

Ruminant livestock animals are a major source of total anthropogenic emissions producing an estimated 80 million tons of CH<sub>4</sub> annually accounting for 33% of anthropogenic emissions of CH<sub>4</sub> (Beauchemin *et al* 2008). Enteric methane is a greenhouse gas that causes significant loses of energy in ruminants and estimated to represent globally 2,079 and 2,344 Mt CO<sub>2</sub>-eq/year for 2010 and 2020, respectively (Hristov *et al.*, 2013). There is therefore an urgent need to develop ways of reducing methane production from ruminants which are major contributors to global warming (CONAM 2001). So, in targeting methane reduction it is crucial to develop a strategy that decrease methane producing micro-biota activities and proliferation without limiting rumen function. Recently there are numerous reports that have shown the reduction of enteric methane due to inclusion of tannin rich browses because the tannins have anti-methanogenic activity, either by direct inhibition of methanogens or indirectly through inhibition of protozoa (Animut *et al.*, 2008; Hristov *et al.*, 2013). Tannins are polyphenolic compounds which bind to protein and can be used as chemical additives for protecting and decreasing ruminant fermentation of proteins in ruminant feeds (Makkar, 2003a). The purpose of the present study was to use an *in vitro* incubation system to screen the potential methane production from a diet based on tannin content of Cassava, *Prosopis juliflora* or their mixture using ammoniated wheat straw as sources of non-protein nitrogen.

## Materials and Methods

### *In vitro* system

An *in vitro* incubation system was used to evaluate the following treatments: (G1): Cassava (C) plus ammoniated wheat straw (Tws) at 50:50. (G2): C plus Prosopis (P) plus Tws at 37.5:37.5:25, respectively. (G3): P plus Tws at 50:50. A representative sample of the mixtures (12 g DM) was subjected to *in vitro* dry matter degradability as described by A.O.A.C (1995).

### Sample collection, preparation and chemical analysis

Samples from each ingredient ((leaves & twigs) were chopped into small pieces and dried in an oven at 55 °C .for 48 h prior to being milled. Feed samples were analyzed for dry matter (DM) and total ash using the method of A.O.A.C (1995). Acid detergent fiber (ADF) and neutral detergent fiber (NDF) were analyzed by the Van Soest method (Van Soest 1965). Gas production was estimated by (Patra *et al.* 2006). Determinations of phenols compound were determined by Maker (2003b). Total phenols and tannins were expressed as tannic acid equivalent and CTs as leucocyanidin equivalent. Blood samples were collected from the jugular vein once before feeding (3 animals in each) at the end of growing period. Blood samples

were centrifuged at 4000 rpm for 20 min. Part of the separated serum was directed to enzymes activity determination, while the other part was stored frozen at -20°C till the biochemical analysis.

### Lamb growth performance

The experiment was conducted in the Borg El Arab Research Station, Animal Production Research Institute, Agricultural Research Center, Egypt. Twenty one growing male lambs of Barki, aged about 3 months and weighed in average  $12.18 \pm 0.17$  kg, divided randomly into three groups, were used to study the effect of the tested rations on growth performance, blood metabolic, feed efficiency. The animals were weighed at the beginning then biweekly. The feeding experiment lasted 16 weeks. The level of the ingredients in the concentrate portion was adjusted to maintain iso-protein and iso-caloric nature in the experimental rations accordingly nutrient requirement (NRC, 1985) for growing sheep.

### Statistical analysis

Data were statistically analyzed using One-Way Layout with Means Comparisons Procedure SAS (2003).

## Results and Discussion

The chemical composition, cell wall constituents and phenols compounds of experimental rations are presented in Table 1. The obtained results showed that the crude protein (CP) was nearly similar in all experimental rations. The highest percent of Crude fiber (CF %), Hemi-cellulose and ADL were recorded with G1. On the other hand, the highest percent of phenols compounds (TP and TT) were recorded with G2.

Table 1. Chemical composition, cell wall constituents and phenols compounds of experimental ratios

Item	Experimental ratios		
	C:Tws (G1)	C:P:Tws (G2)	P:Tws (G3)
DM	80.28	79.50	87.00
Chemical composition:			
OM	85.93	87.23	82.98
CP	15.36	15.40	14.70
CF	48.12	41.23	44.26
EE	3.58	4.22	3.54
NFE	18.87	26.38	20.48
Ash	14.07	12.77	17.02
Fiber fraction % of DM:			
NDF	38.10	38.80	38.70
ADF	19.00	28.00	30.00
Hemi-cellulose*	19.10	10.80	8.70
Cellulose **	9.50	11.20	23.50
ADL	9.50	6.80	6.50
NFC***	28.69	28.81	26.54
NFC/NDF	0.75	0.74	0.69
Phenols compounds g/kg DM:			
TP	39.90	42.28	40.27
TT	16.40	19.2	15.60
CT	20.00	20.00	23.00

\* Hemi-cellulose = NDF-ADF\*\*Cellulose = ADF-ADL

\*\*\*Non fibrous carbohydrates% = OM% - (CP%+NDF%+EE %), Calsamiglia et al., 1995.

Data of methane production are presented in Fig (1). Methane production per unit of substrate fermented was the lowest for G2. This result indicated that methane production showed negative correlation with phenolic compounds (TP and TT). The negative effects of plant phenolic compounds on their fermentation and digestion were reported by Guglielmelli *et al.*, 2011; Jayanegara *et al.*, 2011 and Sebata *et al.*, 2011. The negative effect of tannins on fermentation could be related to the formation of tannin-carbohydrate and tannin-protein complexes that are less degradable or to toxicity to rumen microbes (Bhatta *et al.*, 2009).

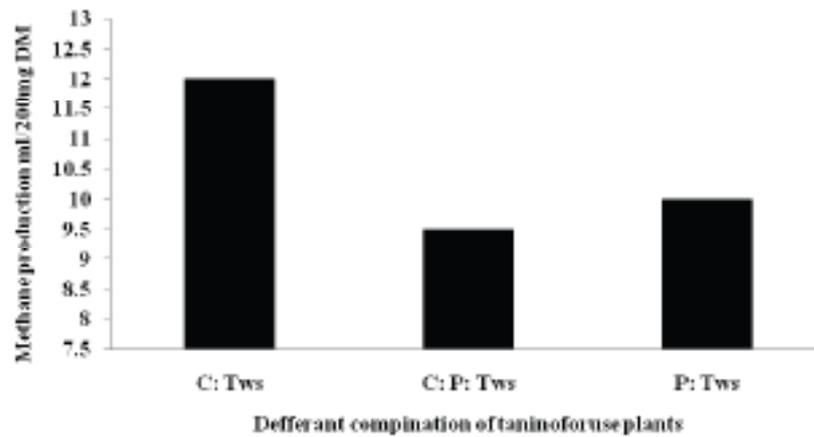


Figure 1. Methane production from the experimental ratios

Table 2 showed that Barki lambs total (TBG) and daily body gains (DBG) were significantly ( $P < 0.05$ ) affected by tested ratios. The highest values were recorded with G2 (22.39 kg and 187 g, respectively).

Table 2. Growth performance and feed efficiency of Barki lambs fed experimental ratios

Item	Groups		
	G1	G2	G3
Initial weight, (kg)	12.20±0.17	12.29±0.33	12.05±0.37
Final weight, (kg)	34.17±0.35 <sup>ab</sup>	34.69±0.40 <sup>a</sup>	33.19±0.45 <sup>b</sup>
Total gain, (kg)	21.97±0.21 <sup>ab</sup>	22.39±0.32 <sup>a</sup>	21.14±0.33 <sup>b</sup>
Daily body gain, (g)	183±1.79 <sup>ab</sup>	187±2.63 <sup>a</sup>	176±2.78 <sup>b</sup>
Total DMI (g/h/d)	734	726	700
Feed efficiency:			
DMI as %BW	3.17	3.09	3.10
DMI g/kg BW <sup>0.75</sup>	69.50	68.09	67.57

a-b Means in the same row with different superscripts differ significantly at  $P < 0.05$ . G1: Concentrate feed mixture (CFM) 40%:Roughage 60% (C plus Tws at 50:50), G2: CFM (40%): Roughage 60% (C plus P plus Tws at 37.5:37.5:25, respectively). G3: CFM (40%): Roughage 60% (P plus Tws at 50:50). CFM consists of 25% undecortecated cotton meal, 43% yellow corn, 25% wheat bran, 3.5% molasses, 2% limestone, 1% common salt and 0.5% minerals mixtures..

Data of blood serum parameters are presented in Table (3). The results indicated that most tested blood parameters (except total protein (TP)) were not significantly affected by tested ratios.

Table 3. Effect of feeding experimental ratios for Barki lambs on some blood serum parameters

Items	Groups		
	G <sub>1</sub>	G <sub>2</sub>	G <sub>3</sub>
Glucose, mg/dl	44.60±0.56	45.84±0.60	44.22±0.43
Total protein, g/dl	6.57±0.78 <sup>ab</sup>	7.57±0.35 <sup>a</sup>	5.17±0.44 <sup>b</sup>
Albumin(A), g/dl	2.96±0.17	3.06±0.07	2.70±0.35
Globulin(G), g/dl	3.61±0.84	3.33±0.44	2.47±0.78
A/G	0.91±0.20	0.95±0.13	1.38±0.45
Urea, g/dl	32.18±0.45 <sup>b</sup>	33.60±0.64 <sup>b</sup>	35.37±0.41 <sup>a</sup>
Creatinine mg/dl	1.17±0.09	1.27±0.09	1.47±0.12
Cholesterol, mg/dl	56.43±1.13	57.53±1.96	55.33±1.20
Triglycerides mg/dl	74.00±1.53	74.33±1.45	73.00±1.22
AST, u/l	32.63±1.23	33.67±1.20	34.07±1.16
ALT, u/l	18.33±1.76	18.33±1.67	18.88±1.69
Calcium, mg /dl	8.49±0.75	8.47±0.77	8.81±0.83
Phosphorus, mg/dl	4.47±0.52	4.50±0.31	4.57±0.61

a-c Means in the same raw with different superscripts differ significantly at  $P < 0.05$ .

## Conclusion

The strategy of mixing between Cassava and Prosopis with ammoniated wheat straw tended to improve animal performance and feeding efficiency by reducing methane emissions per unit of animal product. Under semi-arid area depends on rangeland trees could be solving green fodder shortage especially in summer, season.

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# Effect of Gambir extract (*Uncaria gambir* Roxb) Supplementation as Antioxidant on Performance of ISA-Brown Laying Hens of 40-43 Weeks Old

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## Abstract

*Gambir (Uncaria gambir Roxb) is one of herbs that potential as natural antioxidants. This study aimed to determine the effect of gambir extract supplementation on laying hens performances. The study used a completely randomized design with 3 treatments, 5 replications with 6 hens of each. The variables observed were feed intake, feed conversion ratio (FCR), hen day egg production, egg mass production, blood profile, MDA (Malondealdehyde acid) content of blood plasma and IOFC (Income Over Feed Cost) value. This study used 90 of 40 weeks old Isa Brown laying hens. The treatments were T0 = drinking water without gambir extract, T1 = 20 mg gambir extract /kg BW in the drinking water, T2 = 40 mg gambir extract / kg BW in the drinking water. The results showed that the treatments did not affect the hen performances, blood profile and MDA (Malondealdehyde acid) content of blood plasma. Supplementation of gambir 20 mg/kg BW tend to increase the hen day egg production 5.72%, egg mass production 4.76%, feed efficiency 3.94%, income over feed cost 18.6% and increased antioxidant activity in term of decreasing MDA (Malondealdehyde acid) of the blood and egg. However, supplementation of 20 mg gambir extract / kg BW in the drinking water significantly ( $P < 0.05$ ) increased the eosinofil percentage. Supplementation of gambir extract 20 mg/kg BW or 40 mg/kg BW did not impair the health status of the hens. The conclusion of this study was that gambir extract is potential as source of antioxidant for the laying hens.*

**Keywords:** gambir, laying hens, performance

## Introduction

High ambient temperature in Indonesia (26-31.5°C) as tropical country leads to reduce poultry production due to heat stress. The comfortable zone to optimize laying hen production is 19°C to 22°C (Charles 2002). This heat stress causes oxidative stress to the poultry. According to Mujahid *et al.* (2007), oxidative stress is a condition that free radical activity is higher than antioxidant capacity system. This condition leads to damage unsaturated fat in membrane cell, amino acids of the protein and DNA's nucleotides, and then impairs the membrane and cell integrity (Surai 1999).

Gambir (*Uncaria gambir* Roxb) is one of herbs that potential as natural antioxidants. Gambir extract contains polyphenolic compound such as catechin, tannin, gambirin and saponin as natural antioxidant- and antibiotic. Sahin *et al.* (2010) showed that feeding quails of 5 weeks of old with epigallocatechin-3-gallate extracted from green tea resulted in decreasing of MDA (*malondialdehyde acid*) concentration in the liver from 2.82 to 1.72 nmol g<sup>-1</sup>. MDA (*malondialdehyde acid*) is reactive aldehyde compound produced during peroxidation of poly unsaturated fatty acids in membrane (Vaca *et al.* 1988). According to Hasti *et al.* (2012), feeding mice with gambir extract 30, 100 and 300 mg/kg BW were effective as hepatoprotector. However, usage gambir extract as antioxidant has not been carried out for poultry. The objective of this experiment was to evaluate the effect of gambir extract as source of antioxidant on ISA-Brown laying hen performances.

## Materials and Methods

This study used 90 ISA- Brown laying hens of 40 weeks of old and reared until 43 weeks of old in the battery cages system with the size of 22 cm (width) x 40 cm (length) x 40 cm (height). The composition and nutrients content of the diet used in this study (Table 1) was formulated according to the recommendation of Leeson and Summers (2005). All treatments were fed the same diet. Gambir extract as treatment was administered through drinking water. A completely randomized design using 3 treatments, 5 replications and 6 hens of each was used in this study. The treatments were T0 = drinking water without gambir extract, T1 = 20 mg gambir extract /kg BW in the drinking water, T2 = 40 mg gambir extract /kg BW in the drinking water. The data were analyzed using analyses of variance (ANOVA) according to Steel and Torrie (1995).

Table 1. Composition and nutrients content of the experimental diet

Feed Ingredients	%
Yellow corn	55.00
Rice bran	3.00
Soybean meal	21.00
Meat bone meal	6.00
CPO	4.00
DCP	1.24
CaCO <sub>3</sub>	9.00
Premix	0.50
NaCl	0.16
DL-Methionine	0.10
Total	100.00
Calculated nutrients content:	
Metabolizable energy (kcal/kg)	2877.50
Dry matter (%)	89.74
Ash (%)	5.66
Crude protein (%)	18.15
Crude fiber (%)	2.52
Ether extract (%)	7.04
Ca (%)	4.22
P avail (%)	0.41
Lysine (%)	0.96
Methionine (%)	0.41
Methionine + Cystine (%)	0.68
Na (%)	0.14
Cl (%)	0.19

Table 2. Gambir (*Uncaria gambir* Roxb) extract component

Component	%
Catechin	42.82
Saponin	1.18
Tannin	56.31

Analyzed at Laboratory of Balai Penelitian Tanaman Rempah dan Obat (2014)

## Results and Discussion

### Effects of the Treatments on Laying Hens Performance

The data of laying hen performance obtained in this study are presented in Table 3.

Table 3. The performances of ISA-Brown laying hen during 4 weeks experiment (40-43 weeks old)

Variable	Treatment		
	T0	T1	T2
Feed intake (g/hen/day)	103.39 ± 3.15	105.01 ± 3.20	107.70 ± 2.54
Hen day egg production (%)	75.69 ± 8.50	80.02 ± 6.36	75.99 ± 6.19
Egg mass production (g /4 weeks)	1200.03 ± 71.83	1248.94 ± 49.22	1211.26 ± 77.30
Average egg weight (g/egg)	56.75 ± 3.14	56.16 ± 0.82	59.10 ± 2.46
Feed conversion ratio	2.54 ± 0.33	2.44 ± 0.19	2.52 ± 0.18
IOFC (IDR/hen/4 weeks)	3829.05 ± 2683.11	4540.20 ± 1720.81	3922.22 ± 1734.82

T0 = drinking water without gambir extract, T1 = 20 mg gambir extract /kg BW in drinking water, T2 = 40 mg gambir extract /kg BW in drinking water

The results showed that the treatments did not statistically affect the performance of the laying hen. However, supplementation of gambir extract 20 mg/kg BW (T1) tended to increase egg mass production, feed efficiency, and resulted in increasing income over feed cost with the average value of 4.08%, 3.97%,

and 18.57%, respectively. The hen day production of this experiment achieved 82.3% (T0), 86.98% (T1) and 82.6% (T2) of the standard of ISA-Brown. The average of egg weight of this study achieved 87.7% (T0), 86.8% (T1), and 91.34% (T2) of the standard. According to Hendrix Genetic Company (2011), hen day production of the ISA-Brown laying hens of 40 weeks of old was 92%, and the egg weight was 64.7%. These lower values correspondence with low feed consumption (103.39-107.7 g/hen/day). The recommended feed consumption of ISA-Brown laying hens of 40-43 weeks of old was 115 g/hen/day (Hendrix Genetic Company 2011). The low feed consumption could be associated with high ambient temperature in this experiment. According to Leeson dan Summers (2005), feed consumption was affected by ambient temperature, egg mass production, and energy content of the diet.

Egg production was affected by ambient temperature (Mashaly *et al.* (2004). Egg production of White Leghorns decreased due to high ambient temperature (Kirunda *et al.* 2001). According to Bird *et al.* (2003), high ambient temperature decreased egg production, because lots of energy intake was used to regulate body temperature, in the other hand the feed consumption decreased during this condition. This leads to decreasing of nutrients intake for producing eggs. Supplementation of gambir extract 40 mg/kg BW (T2) tended to increase the egg weight, it could be due to antioxidants activity of gambir extract. According to Bell and Weaver (2002), antioxidants protect fat oxidation and then increased the egg weight. Fat was needed to synthesize the yolk.

### Effect of the Treatments on MDA Concentration of the Blood and Egg of ISA-Brown Laying Hens

The MDA of blood plasma and egg of the laying hens are presented in Table 4.

Table 4. MDA concentration of blood and egg of 43 weeks old laying hens

Variable	Treatment		
	T0	T1	T2
Blood MDA (mmol L <sup>-1</sup> )	6.72 ± 2.17	5.78 ± 2.51	4.88 ± 1.25
Decrease of blood MDA (%)	0	13.99	27.38
Egg MDA (µg/g sampel)	5.67 ± 2.16	5.46 ± 1.01	5.56 ± 1.70
Decrease of egg MDA (%)	0	3.70	1.94

T0 = drinking water without gambir extract, T1 = 20 mg gambir extract /kg BW in drinking water, T2 = 40 mg gambir extract/kg BW in drinking water

The results showed that the treatments did not affect the MDA of blood as well as of eggs. However, these MDA tended to decrease due to the gambir extract supplementation. Supplementation of gambir extract at 40 mg/kg BW decreased MDA 27.38% in the blood of hens. This result indicated antioxidants activity of the gambir extract. Catechin is main compound contained in the gambir extract and has property as antioxidants (Kassim *et al.* 2011). In 1923, Freudenberg and Purman isolated two kinds of active polyphenols (catechin and epicatechin) as source of antioxidants in the dry gambir extract (Nonaka and Nishioka 1980). The gambir extract used in this study contain 42.82% catechin (Table 2).

### Effect of Treatments on Blood Profile of Laying Hens

The blood profile of the laying hens are presented in Table 5.

Table 5. Blood profile of ISA-Brown laying hens of 43 weeks of old

Variable	Treatment			Standard (Swenson 1984)
	T0	T1	T2	
Hemoglobin (g %)	12.84 ± 1.43	11.44 ± 1.24	12.95 ± 1.66	7-13
Hematocrit (%)	22.05 ± 0.39	21.83 ± 1.47	22.44 ± 1.34	22-35
Erythrocyte (10 <sup>6</sup> mm <sup>-3</sup> )	2.50 ± 0.14	2.47 ± 0.32	2.35 ± 0.22	2.5-3.5
Leukocyte (10 <sup>3</sup> mm <sup>-3</sup> )	10.40 ± 4.49	10.80 ± 3.09	10.52 ± 2.52	20-30
Heterophil (%)	36.40 ± 13.43	38.20 ± 16.04	33.60 ± 12.34	25-30
Lymphocyte (%)	56.60 ± 13.76	55.20 ± 15.14	61.40 ± 12.38	55-60
Mesophyl (%)	7.00 ± 1.87	5.80 ± 1.92	5.00 ± 1.00	10
Eosinophil (%)	0.00 ± 0.00b	1.20 ± 0.45a	0.00 ± 0.00b	3-8
HL <sup>-1</sup> (%)	0.76 ± 0.60	0.82 ± 0.61	0.60 ± 0.31	0.45-0.50

T0 = drinking water without gambir extract, T1 = 20 mg gambir extract /kg BW in drinking water, T2 = 40 mg gambir extract /kg BW in drinking water

The results showed that supplementation of gambir extract at 20 mg/kg BW significantly increased ( $P<0.05$ ) eosinophil in the blood of hens. This result indicated that active compound in the gambir extract such as catechin increases immune response of hens. Eosinophil helps to protect the body against disease and infections by moving around and eating some types of bacteria, foreign substances, and other cells. The treatments did not affect the other blood profile variables observed in this experiment. This condition indicated that gambir extract did not impair the health status of the hens.

## Conclusion

Gambir extract with high content of catechins was potential as source of antioxidants for laying hens. Gambir extract decreased MDA (Malondialdehyde acid) content of blood plasma, increased immune response and did not impair the health status of hens.

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# Root Tubers as Alternative Energy Sources in Rabbit Ration: Effect on Growth Performance and Economic Value

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## Abstract

Rabbit needs more energy to growth on weaning periods. Various kinds of root tubers can be used as feed energy sources but there are limited usage information about them. This study evaluated the growth performance and economic value of rabbit fed different kinds of root tubers as energy sources in the ration. Completely randomized design was used on this study. Twenty male rabbits on weaning period were divided into 4 feeding treatments with different kinds of root tubers, namely: R1: corn (control), R2: sweet potatoes, R3: cassava, R4: taro. The root tubers were used as much as 30% on the rations. The observed parameters were dry matter intake (DMI), water consumption, weight gain, feed efficiency (FE), feed conversion (FCR) and economic value. The results showed that different kinds of root tubers had significantly affected DMI. Sweet potatoes and cassava as feed energy sources had produced the lowest DMI ( $P < 0.05$ ). However the treatments did not affect in water consumption, weight gain, FE and FCR. The use of taro gave the best economic value than others. It was concluded that the use of sweet potatoes, cassava and taro at the level of 30% can be used as alternative energy sources on rabbit ration.

Keywords: energy sources, performance, rabbit, root tubers

## Introduction

The nutrient requirement of rabbit on the weaning periods is higher than the adults. Nowadays, corn is commonly used as an energy source on rabbit ration though it is expensive, highly competed with human as food and agro-industries. The other feedstuffs are needed to replace corn as the energy source. Sweet potatoes, cassava and taro are root tubers which have a large potential production (BPS, 2015). Sweet potatoes (*Ipomoea batatas* (L.)) is a good energy source feedstuff because it contains of easily digestible carbohydrate (Lingga *et al.*, 1989). Cassava (*Manihot utilissima* Pohl) as the carbohydrate source, contains 64-72% of starch, and almost 99% of it was amylose and amylopectin (Davendra, 1977). The major factor against the use of cassava root is the cyanide (HCN) content which affects the nutrient utilization in animals. Agunbiade *et al.*, (2002) reported that HCN content on cassava peel meal is 27 mg/kg. Cyanide toxicity can lead to increase the respiration rate, pulse rate and spasmodic muscular movement (Oke, 1969). Taro (*Xanthosomasagittifolium* (L.)) contains digestible energy as much as 3474 kcal/kg (Departemen Kesehatan RI, 1955). Taro roots was an ideal feedstuff as the carbohydrate sources (Lingga *et al.*, 1989). There is limited information on the use of three kinds of root tubers in the rabbit ration. Therefore, this study evaluated the growth performance and economic value of rabbit fed different kinds of root tubers as energy sources in the ration.

## Materials and Methods

### Animal and Diets

A total of twenty male rabbits weaned at 4-5 weeks with a mean initial body weight of  $271 \pm 32$  g were randomly allotted into 4 feeding treatments (5 replications of each). The rabbits were housed individually equipped with feeder and drinker in cage. The dietary treatments consisted of 4 rations with different kinds of root tubers, namely: R1: corn, R2: sweet potatoes, R3: cassava, R4: taro. The root tubers were used as much as 30% on the rations. The diets were formulated on iso calorie and iso protein according to NRC (1977) (Table 1).

Table 1. Ingredients and chemical composition of dietary treatments

Ingredients	R1	R2	R3	R4
	%			
Soybean meal	20	20	20	20
Rice bran	7	7	7	7
Leaves + stems of sweet potato	20	20	20	20
Nativegrass	20	20	20	20
Corn	30	0	0	0
Sweet potato	0	30	0	0
Cassava	0	0	30	0
Taro	0	0	0	30
Crude palm oil	2	2	2	2
Premix	0.5	0.5	0.5	0.5
CaCO <sub>3</sub>	0.5	0.5	0.5	0.5
Total	100.0	100.0	100.0	100.0
Nutrient composition (DM)				
Dry matter (%)	87	87	87	86
Crude fiber (%)	14	17	14	15
Crude protein (%)	15	14	14	14
Crude fat (%)	6	5	5	5
Ash (%)	10	10	9	10
Ca 9%)	2	1	2	2
P (%)	0.9	0.8	0.8	0.8
NFE (%)	56	53	57	55
GE (Kcal/kg)	4760	4289	4330	4163
DE* (Kcal/kg)	3708	3663	3670	3653

Note: \* Calculated values; DM: dry matter, NFE: nitrogen free extract, ADF: acid-detergent fibre, GE: gross energy, DE: digestible energy.

### Pellet Processing

Root tubers were sliced with slicer. Sweet potato leaves and stems as well as native grass were chopped. Feed ingredients were sun dried for half day, then they were dried using 105°C on oven for 15 minutes. Feed ingredients were grinded and formulated. The mixed ration were formed pellet on pellet machine.

### Experimental Procedure

The experiment lasted 56 d and during that time, feed and water were offered *ad-libitum*. The difference between quantity fed and left over gave the consumed quantity of rabbit. Weight gained, DMI and water consumption were determined weekly. FE and economic value were determined at the end of feeding trial. Procedures of AOAC (2000) were used to determine proximate composition, calcium (Ca) and phosphorus (P) of dietary treatments. Gross energy (GE) of the diets was determined using bomb calorimeter.

Data were analyzed by the one way analysis of variance (ANOVA). Differences between mean values were separated using Duncan's Multiple Range Test.

### Results and Discussion

The results showed that DMI was significantly influenced by different kind of root tubers ( $P < 0.05$ ), but water consumption, weight gain, FE as well as FCR were not significantly affected by the treatments (Table 1). Taro as energy sources had the highest DMI than others. Sweet potatoes and cassava resulted the opposite of those results. The amount of energy of root tubers affected DMI. The lowest digestible energy of taro treatment causing the highest DMI. Cheeke (1987) reported that DMI will increase if the energy ration is low. This study showed the similar water consumption among the treatments, but the total of water consumption was higher than Church (1991) who stated that rabbit water consumption was 135-150 mL/day at the age of 5-10 weeks. The energy content on the rations was higher than energy requirement which are 2500 kcal/kg (NRC, 1977). The increase of environment heat will cause the increase of water consumption. Taro treatment had the highest weight gain, but that is not significantly different than others. Weight gain on this study was 16-20 g/day. It was higher than Sunarwati (2001) who reported that the weight gain of rabbit fed by sweet potatoes biomass was 8.8-18.4 g/day.

Table 2. The effect of root tubers treatments on dry matter intake, water consumption, daily weight gain, feed efficiency, feed conversion ratio

	Treatments			
	R1	R2	R3	R4
DMI (g/day)	61 ± 9.7 <sup>ab</sup>	56 ± 5.4 <sup>b</sup>	57 ± 6.5 <sup>b</sup>	69 ± 6.5 <sup>a</sup>
Water consumption (mL/day)	189 ± 31	155 ± 22	160 ± 47	178 ± 22
Weight gain (g/day)	20 ± 3	16 ± 4	17 ± 4	21 ± 3
FE	0.29 ± 0.01	0.25 ± 0.05	0.25 ± 0.04	0.26 ± 0.04
FCR	3.48 ± 0.15	4.07 ± 0.82	4.06 ± 0.75	3.87 ± 0.53

Note : <sup>ab</sup>Mean with different superscript within the same raw are different (P<0.05); R1: corn, R2: sweet potatoes, R3: cassava, R4: taro; DMI : dry matter intake, FE : feed efficiency, FCR: feed conversion.

FE and FCR also did not significantly affected by different kind of root tubers in the ration. Taro treatment had better FE and FCR than other kinds of root tubers. However this result is still lower than corn treatment. It was in accordance with Cheeke (1987) who stated energy content in the ration will be affected on FE and FCR values.

Table 3. The effect of root tubers treatments on economic value

	Treatments			
	R1	R2	R3	R4
Total income	24,000	20,000	22,000	26,000
Total expense	15,971	15,203	14,936	17,113
Profit	8,029	4,797	7,064	8,887
Feed price	2,059	1,927	1,828	2,092

Note : R1: corn, R2: sweet potatoes, R3: cassava, R4: taro

Table 3 showed that different kind of root tubers was given different result on economic value. The highest profit was obtained on taro treatment, but the feed price was also higher in this treatment. Cassava treatment was provided the lowest price. It caused by the different price of root tubers. Taro price was higher due to uncommon used as feed ingredient, but cassava price was cheaper.

## Conclusion

The use of sweet potatoes, cassava and taro at the level of 30% can be used as alternative energy sources for rabbit ration.

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# Live Weight Gain of Beef Cattle Fed on Complete Feed Silage of Water Hyacinth Supplemented with Mineral Zinc-Proteinate

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## Abstract

Beef cattle business development is often constrained by the supply of feed. Weed water hyacinth potential as feed for cattle. The aim of this study was to assess the provision of complete feed silage of water hyacinth (CFSWH) and supplementation with mineral Zn-proteinate on growth of beef cattle. CFSWH made from a mixture of fresh water hyacinth and concentrate with a comparison of 2:1, then stored in silo for 2 weeks. The experimental design used was completely randomized design 4x4 with a cross-over pattern. Eight Freisian Holstein steers maintained for 8 months, divided into two periods for each period of 4 months. Four ration treatment (PK 11%, 62% TDN) was: P0= elephant grass+ concentrate (control), P1= 50% control+50% CFSWH, P2= 100% CFSWH, P3= 100% CFSWH+Zn-proteinate 40 ppm. The results showed that the quality of CFSWH was good based on pH and nutrient content. Dry matter intake in the first periode (first 4 months) was not significantly different between treatments, but in the second period giving CFSWH 100% (P2) was significantly reduced consumption of DM ( $P < 0.05$ ). Live weight gain (LWG) and feed conversion were not significantly different in overall period, but the second period resulted in the LWG higher than the first period (1.3 vs 0.88 kg/day). Supplementation of Zn-proteinate on CFSWH (P3) resulted in the LWG higher than without supplementation (P2) in the first period. The conclusion of this study is complete feed silage of water hyacinth can replace conventional ration in beef cattle business, Zn-proteinate supplementation is needed in the initial period of growth.

Keywords: cattle, complete feed, live weight gain, water hyacinth, zinc

## Introduction

Beef cattle business development is often constrained by the supply of feed. Weed water hyacinth potential as feed for cattle, besides having lots of fiber cellulose and hemicellulose, this plant also has a very high nutrient content. Mako *et al.* (2011), which examined the nutrient content of different water hyacinth (canals, lakes, rivers, dams) get a crude protein content of 10.4%; 18.7% crude fiber; Neutral Detergent Fiber (NDF) from 65.9 to 77.9%, and the levels of Acid Detergent Fiber (ADF) from 36.5 to 39.7%. However, the water content and high oxalates made water hyacinth cannot be given to cattle in fresh form. Processing water hyacinth into complete feed silage is an effort to improve its utilization as animal feed. The provision of complete feed silage of water hyacinth as feed on the sheep have been tried and resulted in body weight gain were quite good (Muktiani *et al.*, 2015).

Mineral supplementation Zn (Zinc) is necessary given because very limited in the feed. The content of zinc in ruminant feed in Indonesia generally range between 20-38 mg / kg DM (Adriani *et al.*, 2004), whereas it should contain a minimum of 30 mg / kg to feed beef cattle (NRC, 1996). Fu-yu *et al.* (2007) explains that the mineral zinc is a trace mineral that is a constituent of the 300 kinds of enzymes associated with the metabolism of proteins, amino acids, nucleic acids, fats, carbohydrates and vitamins. The aim of this study was to assess the provision of complete feed silage of water hyacinth (CFSWH) and supplementation with mineral Zn-proteinate on growth of beef cattle and feed conversion.

## Materials and Methods

Complete feed silage of water hyacinth (CFSWH) made from a mixture of fresh water hyacinth and concentrate with a comparison of 2:1. Concentrate containing 66% Total Digestible Nutrients (TDN) and 13% crude protein, composed of 14.5% cassava waste, 31% rice bran, 13% palm kernel meal, 8% coffee leather, 27.5% coconut meal, 3.5% kapok seed meal and 2.5% molasses. Fresh water hyacinth reduced

water content with drained for 24 hours. Furthermore, chopped 3-5 cm and mixed with the concentrate and stored in silos for 2 weeks.

The experimental design used was completely randomized design 4x4 with a cross-over pattern. Eight Freisian Holsteinstears maintained for 8 months, divided into two periods for each period of 4 months. Four ration treatment (PK 11%, 62% TDN) was: P0= elephant grass+concentrate (control), P1= 50% control+50% CFSWH, P2 = 100% CFSWH, P3= 100% CFSWH+Zn-proteinat 40 ppm. The content of feed ingredients used in this study are listed in Table 1.

Table 1. The nutrient content of the feed materials research

Nutrient	Elephant grass	Water hyacinth	Concentrate
Dry matter (%)	17.94	8.26	88.47
Total Digestible Nutrient (%)	48.50	41.79	66.08
Crude protein (%)	11.56	10.52	13.14
Ash (%)	14.94	17.02	12.62
Fat (%)	4.73	0.87	4.76
Crude Fiber (%)	42.7	45.99	22.83
Nitrogen Free Extract (%)	20.07	25.59	46.65

Parameters measured were quality of silage (pH and nutrient content), feed consumption and weight gain were measured boot every 2 weeks. The data were analyzed with analysis of variance, if any effect of treatment followed by Duncan's multiple range test (Steel and Torry 1981).

## Results and Discussion

Based on nutrient and pH silage, CFSWH can be classified a good quality that has a pH of 4.27. Ratnakomala (2006) stated that pH of good silage range between 3.8 to 4.2. Content of dry matter was decreased from 41.23% to 39.29% (lost 1.94%) and crude protein decreased 11.88% to 10.78% (lost 1.10%) after stored for two week. Dry matter loss is an indicator of nutrient degradation during the process ensilage. Muck (2011) argues that the loss of 1-10% dry matter after ensilage process can still be tolerated. Protein content of CFSWH was sufficient to yielded average daily gain (ADG) between 0.8 to 1.2 kg/day (NRC, 1996). Dry matter intake, body weight gain and feed conversion are presented in Table 2.

Table 2. Dry matter intake, body weight gain and feed conversion in treatment cattle

Parameters	Treatment				Average
	P0	P1	P2	P3	
Period I					
Dry matter intake (Kg)	8.26	8.32	6.31	8.71	7.78
Dry matter intake(% LW)	2.96	2.88	2.47	2.95	2.8
Period II					
Dry matter intake (Kg)	9.85	9.20	9.90	9.10	9.46
Dry matter intake(% LW)	2.72 <sup>a</sup>	2.62 <sup>a</sup>	2.49 <sup>b</sup>	2.77 <sup>a</sup>	
Average					
Dry matter intake (Kg)	9.05 <sup>a</sup>	8.76 <sup>a</sup>	7.30 <sup>b</sup>	7.76 <sup>b</sup>	
Dry matter intake(% LW)	2.84 <sup>a</sup>	2.75 <sup>a</sup>	2.48 <sup>b</sup>	2.86 <sup>a</sup>	

Note: P0= elephant grass+concentrate (control), P1= 50% control+50% CFSWH, P2= 100% CFSWH, P3= 100% CFSWH+Zn-proteinat 40 ppm.

Cows were fed on complete feed silage of water hyacinth showed consumption significant ( $P < 0.05$ ) lower than the ration of grass and concentrates. Dry matter intake in the first periode (first 4 months) was not significantly different between treatments, but in the second period giving CFSWH 100% (P2) was significantly reduced consumption of DM ( $P < 0.05$ ). Supplementation of mineral Zn proteinat can increase dry matter intake equal the control diet. However, the level of consumption of cattle fed complete feed silage hyacinth pretty good, so it can certainly needs adequate nutrients, if calculated based percent body weight, dry matter intake was normal, around 2.2-3% (NRC, 1996).

The average daily weight gain and feed conversion calculation to the cows experiment is presented in Table 3.

Tabel 3. Average daily gain (ADG) and feed conversion

Parameters	Treatment				
	P0	P1	P2	P3	Average
Period I					
ADG (Kg/hari)	0.94	1.01	0.58	1.07	0.88
Feed conversion	8.95	8.21	11.00	8.13	9.20
Period II					
ADG (Kg/hari)	1.16	0.90	1.32	0.87	1.30
Feed conversion	9.05	10.28	7.49	10.63	9.63
Average					
ADG (Kg/hari)	1.05	0.96	0.83	0.94	
Feed conversion	8.60	9.14	8.84	8.29	

Note: P0= elephant grass+concentrate (control), P1= 50% control+50% CFSWH, P2= 100% CFSWH, P3= 100% CFSWH+Zn-proteinate 40 ppm.

The feeding of CFSWH produce ADG were not significantly different in overoll period compared to conventional ration which ranged from 0.83 to 1,05 kg/day. The average ADG of cattle in the second period was higher than the first period (1.3 vs. 0.88 kg/day), but also higher in feed conversion. Daily body weight gain of cattle were given CFSWH (without grass) was good, this indicated that CFSWH can replace conventional rations. Supplementation of Zn-proteinate on CFSWH (P3) resulted in the LWG higher than without supplementation (P2) in the first period Fu-yu et al. (2007 ) explains that the mineral zinc is a constituent of the 300 kinds of enzymes associated with the metabolism of nutrient, so impact on the increased of nutrients utility and body weight gain. Feed conversion in this study (8.29-9.14) can be classified in the good. The research of O'Kiely (2011) get feed conversion was 7.14 to 12.15 on cattle fed different type of silage.

## Conclusion

The conclusion of this study is complete feed silage of water hyacinth can replace conventional ration in beef cattle business, Zn-proteinate supplementation is needed in the initial period of growth.

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# The Effect of Adding Fermented Waste Cabbage in Calf Starter Pellets on Total Lactic Acid Bacteria and *Escherichia coli*

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## Abstract

When calf is first born, its rumen is sterile because are no bacteria present. Calf starter is then used as a link to rumen development and successful weaning. However, young calves tend to easily catch diarrhea. Therefore, adding fermented waste cabbage on calf starter pellets is needed in order to rumen development, to improve calf immunity and to reduce its potency to suffer from diarrhea. This research aimed to examine the effect of addition microbial source of fermented waste cabbage in calf starter pellets on total lactic acid bacteria and the presence of *Escherichia coli*. The materials of calf starter were corn, soybean meal and bran, molasses, mineral mix and materials in fermented waste cabbage from cabbage waste, sugar and salt. Research uses completely randomized design with 4 treatments and 5 replications (T0: 0% fermented waste cabbage + 100% calf starter, T1: 2% fermented waste cabbage + 100% calf starter, T2: 4% fermented waste cabbage + 100% calf starter, T3: 6% fermented waste cabbage + 100% calf starter). The parameters measured were total lactic acid bacteria and *Escherichia coli*. The data were analyzed with descriptive analyze. Results showed that the more addition of fermented waste cabbage, the higher the count of lactic acid bacteria (T0:  $3,3 \times 10^3$  cfu/g; T1:  $6,0 \times 10^5$  cfu/g; T2:  $6,3 \times 10^3$  cfu/g; T3:  $8,0 \times 10^6$  cfu/g) and there were no bacteria *Escherichia coli* present on calf starter pellets.

Key words: calf starter, *Escherichia coli*, total lactic acid bacteria

## Introduction

Calves at birth have very different digestive tract from adult cattle. New born calf's rumen is still sterile and undeveloped since there are no bacteria present (Quigley 2001). Feeding calf starter can stimulate rumen development and is best given to 2 – 6 week-old calves (Cunningham 1995). It will accelerate the process of weaning calves as well.

Another problematic issue concerning young calves is that they tend to catch diarrhea easily. According to Subronto's (2004) research, newborn calves' (aged 2-10 days) mortality rate caused by diarrhea is 10 - 50%. Diarrhea is generally caused by *Escherichia coli* (Fardiaz 1993). Dairy farmers often use antibiotics for diarrhea treatment. However, based on the patterns of resistance, antibiotics can be rendered ineffective.

Probiotic can be used alternatively as natural antibiotics. Probiotics are living non-pathogenic organisms capable of maintaining the balance of intestinal micro flora in the digestive tract (Shitandi *et al.* 2007). Feeding calves with probiotic-rich pellets can optimize calves' growth by reducing their potential to catch diarrhea. Fermented waste cabbage is selected as the source of probiotics.

Waste cabbage is by product of cabbage's outer shells that have been sorted. Waste cabbage naturally contains lactic acid bacteria and fermentation process can increase the number of lactic acid bacteria. The addition of microbial lactic acid bacteria from fermented waste cabbage can enhance calf starter's benefits.

The research aimed to examine the effect of adding a source of microbial fermented cabbage waste in calf starter pellets on total lactic acid bacteria and the presence of the bacterium *Escherichia coli*.

## Materials and Methods

The research material included materials and equipment. Corn, rice bran, soybean meal, molasses, mineral mix, waste cabbage, sugar, salt, de Man Rogosa Sharpe (MRS) medium, Tryptic Soy Broth medium (TSB) and Eosin Methylene Blue Agar (EMBA) medium were the required materials. Equipment used were knives, digital scales, trays, plastic, tape, labels paper, pelleter machine, stove, boiler, digital pH meter, plastic wrapping, oven, incubator, autoclave, measuring cups, sterile petri dish, pipette 1 ml, tube test, spatula, erlenmeyer and quebec colony counter.

The research used completely randomized design with 4 treatments and 5 replications (T0: 0% fermented waste cabbage + 100% calf starter, T1: 2% fermented waste cabbage + 100% calf starter, T2: 4% fermented waste cabbage + 100% calf starter, T3: 6% fermented waste cabbage + 100% calf starter).

The method consisted of two stages. The first stage was fermenting waste cabbage. Waste cabbage was cut into small pieces, blended and added 6% salt and 6.4% sugar and then fermented in anaerobic condition for 6 days. The second stage was making the pellets. Formula calf starter contained 19.62% crude protein and TDN 79.41% (Mukodiningsih *et al.* 2010). After all the ingredients were mixed, calf starter went through conditioning process at 80°C temperature for 20 minutes. Before extruding process, the temperature of calf starter should be decreased at 30°C temperature and then fermented waste cabbage was added. Pellets were extruded with diameter sized 5 mm. Then, pellets were dried in oven at 34-39°C temperature to reach 13% water content.

The parameters observed were total lactic acid bacteria and *Escherichia coli*. The data of total lactic acid bacteria and *Escherichia coli* were analyzed using descriptive analysis (Belanche *et al.* 2011).

## Results

Total lactic acid bacteria and *Escherichia coli* in calf starter pellets during research were shown in Table 1.

Table 1. Mean of total lactic acid bacteria, *Escherichia coli* of calf starter pellets with addition of microbial source from fermented waste cabbage

Treatment	Total Lactic Acid Bacteria	<i>Escherichia coli</i>
	-----cfu/g-----	
T0 (0%)	3.3x10 <sup>5</sup>	-
T1 (2%)	6.0x10 <sup>5</sup>	-
T2 (4%)	6.3x10 <sup>5</sup>	-
T3 (6%)	8.0x10 <sup>6</sup>	-

### Total Lactic Acid Bacteria

The treatment of adding microbial source from fermented waste cabbage in calf starter pellets revealed that the increase of fermented waste cabbage addition had consequently increased total lactic acid bacteria (T0: 3.3x10<sup>5</sup>cfu/g; T1: 6.0x10<sup>5</sup>cfu/g; T2: 6.3x10<sup>5</sup>cfu/g; T3: 8.0x10<sup>6</sup>cfu/g). It showed that lactic acid bacteria could grow and developed properly since the environmental conditions were favorable for them. Lactic acid bacteria's growth was influenced by several factors including: temperature, pH, and nutrient sources (the amount of fermentable carbohydrates or sugars). The addition of molasses in calf starter ration and sugar in fermented waste cabbage was adequate for lactic acid bacteria's nutrient requirement. Molasses contained glucose, carbohydrate and organic acid. Molasses and sugar functioned as carbon and nitrogen sources that were used as energy for bacteria (Richana *et al.* 2000). Microorganism especially lactic acid bacteria required nutrients such as carbohydrates for survival (Supardi & Soekamto 1999).

Regarding its acidity, calf starter pellet's pH ranged from 5.50 to 5.76. These conditions are optimal conditions for the growth of lactic acid bacteria. The optimal temperature for lactic acid bacteria was 25-37°C (Supardi & Soekamto 1999). Lactic acid bacteria can grow in pH ranged from 3 to 10.5, but the optimal pH range was 5.5-6.0 (Jay 1996).

### *Escherichia coli*

The results of microbiological analysis showed that *Escherichia coli* were not found in calf starter pellets. *Escherichia coli* grew optimally at pH range 7.0 - 7.5. The pH of pellets range 5.50 - 5.76. This acidic condition was considered the deciding factor *Escherichia coli* could not grow. Acidic condition inhibited the growth of some types of pathogenic microorganisms. Acidic condition in pollard extract fermented with vegetable waste inhibit gram-negative bacteria such as *Salmonella* and *Escherichia coli* to grow, so that the dominant population of microorganism were gram-positive bacteria (Utama *et al.* 2013).

*Escherichia coli* lived in the temperature ranged from 10 to 40°C with an optimum temperature of 37°C. Conditioning process in pellets industry was 80°C during steaming. This condition was lethal for pathogens in feed material. This was in accordance with Parker (1988) which stated that the process of conditioning that aimed to gelatinized also served to reduce the number of gram-negative bacteria or pathogens that might be present in the feed material in pellets production.

Lactic acid bacteria were the reason why *Escherichia coli* was not found. Lactic acid bacteria produced hydrogen peroxide that inhibit decomposing microorganisms through oxidation effect on microorganism's

cell membranes. Hydrogen peroxide's activity could damage the cytoplasmic membrane of gram-negative bacteria while gram-positive bacteria were able to survive because of the structure of cell membrane is thicker than the gram-negative bacteria (Muwakhid *et al.* 2007). Furthermore, Lunggani (2007) added that lactic acid bacteria were capable of inhibiting gram-negative bacteria.

## Conclusion

In conclusions, the higher addition of fermented waste cabbage the better increasing of total lactic acid bacteria. Moreover *Escherichia coli* were not found in the pellets product. Biological analysis needed to be held to determine the effect of pellets calf starter enrich with lactic acid bacteria against calves diarrhea.

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# Substitution of Fish Meal by Cricket or Indigofera Shoot Leaf Meal on Laying Japanese Quail (*Coturnix japonica*) Performance

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## Abstract

Cricket meal and indigofera shoot leaf meal are animal and vegetable feed ingredients which can be used as a source of protein for poultry and can substitute fish meal. Cricket meal contains 55.59% crude protein, 9.71% crude fiber, 14.67% crude fat and 5009 kcal/kg gross energy, meanwhile Indigofera shoot leaf meal contains 28.98% crude protein, 19.96% crude fiber, 9.96% crude fat, and 4253 kcal/kg gross energy. This experiment was carried out to study the effect of fish meal substitution by cricket and indigofera shoot leaf meal in the diet on quail (*Coturnix japonica*) performance. The treatments were R0 (ration contained fish meal), R1 = substitution of fish meal with cricket meal, R2 = substitution of fish meal with Indigofera shoot leaf meal, based on dry matter requirement. Variables measured were feed intake, protein intake (Pi), crude fiber intake (Cfi), fat intake (Fi), calcium intake (Ca), feed conversion ratio (FCR), egg production, egg weight, egg mass production, and hen day. The result showed that feed intake in R1 was higher than other treatments. The fat intake, hen day, and egg mass production were also higher in R1 compared to other treatments ( $P < 0.05$ ), on the other hand crude fiber intake was higher in R2 than other treatments, meanwhile calcium (Ca) intake, feed conversion ratio (FCR), egg production, and egg weight were same in all treatments. This indicated that Cricket meal and indigofera shoot leaf meal can be used as an alternative protein source to substitute fish meal in quail diet.

Keyword : cricket meal, fish meal, indigofera shoot leaf meal, performance, Quail

## Introduction

*Coturnix japonica* is one type of poultry-producing eggs with a fast production cycle. Tuleun and Dashe (2010) reported that quail mature sex around the age of six weeks and generally reach peak production of eggs after 50 days. Nutritional requirement of quail phase-laying is 20% crude protein, 2900 kcal/kg feed energy metabolism (EM), and 1% fat (NRC 1994). The process of egg production is determined by the sufficiency of nutrient from diet. Protein is one of the macro nutrients that affect egg production. In general fish meal, soybean meal, MBM used as protein sources on diet and availability of feed ingredients are imported. Therefore we need an effort to find alternative feed ingredients. The feed ingredients that can be used are *Indigofera zollingeriana* and crickets. *Indigofera zollingeriana* is a leguminosa which is usually used as feed ingredients for ruminants. Poultry intolerance with feed ingredients that have high crude fiber, so that part of the plant used is shoot of the plant because it has crude fiber lower than other part of plant. Indigofera shoot leaf meal contains 28.98% crude protein, 19.96% crude fiber, 9.96% crude fat, and 4253 kcal/kg gross energy. In addition *Indigofera zollingeriana* had  $\beta$ -carotene as a precursor of vitamin A so that can be increased vitamin A on egg. Crickets can be used as a feed source of protein as a substitution for soybean meal on laying hens (De Foliar *et al.* 1982). Poultry intolerant of the feed materials that have a high part of chitin crickets used is the abdomen. Cricket meal contains 58.3% crude protein, 9.71% crude fiber, 10.3% crude fat and 5009 kcal/kg gross energy (Wang 2005). Based on the potential of *Indigofera zollingeriana* and crickets can be used as a substitution of fish meal and it is expected to improve the performance of quail (*Coturnix japonica*).

## Materials and Methods

The treatments were T0 = control (8% ration contained fish meal), T1 = substitution of fish meal with cricket meal 8%, T2 = substitution of fish meal with Indigofera shoot leaf meal 15%, based on dry matter requirement (Table 1).

Table 1. Gross Composition of Experimental Diets

Ingredients	Composition (%)		
	T0	T1	T2
Corn	51.95	51.95	49.04
Rice Bran	4.5	2.95	-
Soybean Meal	23	23	22.5
Fish Meal	8	-	-
Cricket Meal	-	8	-
Indigofera Shoot Leaf Meal	-	-	15
Palm oil	4.5	4.5	4
DCP	0.99	1.5	1.5
CaCO <sub>3</sub>	6.45	7.2	7.2
Nacl	0.25	0.4	0.4
Premix	0.2	0.2	0.2
DL-Methionine	0.16	0.3	0.16

Thirty of female Japanese quails (*Coturnix coturnix japonica*) (6 weeks old with av. BW 150 g) were divided into three treatment (10 per treatment). The treatments were T0= control ( 8% ration contained fish meal), T1= substitution of fish meal with cricket meal 8%, T2= substitution of fish meal with Indigofera shoot leaf meal 15%, based on dry matter requirement. The quails were housed in cage with size 60 cm x 40 cm x 20 cm. Cricket meal contained only abdomen without chitin. The quails were weighed on the first day before the diets were administered. Variables measured were feed intake, protein intake (Pi), crude fiber intake (Cfi) , fat intake (Fi), calcium intake (Ca), feed conversion ratio (FCR), egg production, egg weight, and hen day.

## Results and Discussion

Result showed that during one month feeding trial, feed intake in T1 was higher than other treatments ( $P<0.06$ ). The protein intake, fat intake, egg mass production and hen day were also higher in T1 compare to other treatments ( $P<0.05$ ), on the other hand crude fiber intake was higher in R2 than other treatments, meanwhile calcium (Ca) intake, feed conversion ratio (FCR), and egg weight were same in all treatments.

Table 2. Effect of Substitution of Fish Meal by Cricket or Indigofera Shoot Leaf Meal on Japanese Quail (*Coturnix japonica*) Performance

Parameter	Treatment		
	T0	T1	T2
Feed intake (g/d)	17.80±1.01 <sup>AB</sup>	18.86±0.82 <sup>A</sup>	17.55±0.52 <sup>B</sup>
Protein intake (g/d)	3.323 ± 0.223 <sup>ab</sup>	3.524 ± 0.199 <sup>a</sup>	3.143 ± 0.075 <sup>b</sup>
Crude fiber intake (g/d)	0.46±0.03 <sup>c</sup>	0.59±0.03 <sup>b</sup>	0.86±0.03 <sup>a</sup>
Fat intake (g/d)	1.24±0.07 <sup>b</sup>	1.49±0.07 <sup>a</sup>	1.31±0.04 <sup>b</sup>
Calcium intake (g/d)	0.58±0.03	0.58±0.03	0.55±0.02
FCR	2.04±0,13	2.14±0,16	1.96±0,12
Egg weight (g)	8.75±0,37	8.86±0.59	8.96±0.44
Hen day	60±12.78 <sup>ab</sup>	74.64±18.46 <sup>a</sup>	53.21±19.18 <sup>b</sup>

<sup>A, B, AB</sup>Different superscript in the same line means significantly different ( $P<0.06$ )

<sup>a, b, ab</sup>Different superscript in the same line means significantly different ( $P<0.05$ )

T0= control (ration contained fish meal); T1= substitution of fish meal with cricket meal; T2= substitution of fish meal with Indigofera shoot leaf meal

The performance of Japanese quail fed cricket or Indigofera shoot leaf meal as a substitution of fish meal is shown in Table 1. Japanese quail fed cricket meal have higher ( $P<0.06$ ) feed intake than control dan Indigofera shoot leaf meal. Higher palatability of cricket meal as compared to control/ fish meal and Indigofera shoot leaf meal might be the reason for having higher feed intake by Japanese quail fed with cricket meal. Cricket meal also increase protein intake ( $P<0.05$ ), cricket has high contain of crude protein 55,59%. Likewise fat intake on cricket diet was higher than fish meal and indigofera shoot leaf meal,

because cricket has high of crude fat (10.3%) with the percentage of fatty acid are palmitate (16:0) 50.32%, stearate (18:0) 32.06%, oleat 9.77%, and linoleat 2.34% (Chakravorty *et al.* 2014).

Diet Indigofera shoot leaf meal significantly higher ( $P<0.05$ ) in crude fiber intake than diet control and cricket meal. This may be mainly due to Indigofera shoot leaf meal having higher crude fiber 19.96% than fish meal and cricket meal. Japanese quail fed cricket meal have higher crude fiber intake than Japanese quail fed control, this may be mainly due to cricket meal still contains chitin.

The result of feed conversion ratio (FCR) showed that diet was not significant. In favour of diet Cricket meal (T1) with the highest value while the least was found in diet Indigofera shoot leaf meal (T2). Both whole Fish meal (T0) and indigofera shoot leaf meal (T2) i.e diets T0 and T2 have a lower ( $P<0.05$ ) cost per kilogram of feed as compared with Cricket meal (T1). With FCR results, indigofera shoot leaf meal (T2) requires less cost to produce the diets and still give a good performance. The value of FCR was influenced by feed intake and egg mass production. Influenced FCR by diet because the quail in finish periode production and not top periode production. This was agreement with the findings of Ensminger (1992) that FCR influenced by factor strain, age, management, disease and diet. Value of FCR meaning to diet quality, diet quality is good was value of FCR is low. Good quality of diet influenced by nutrient of balance in diet. Zuprizal (1998) and Sunafik (2000) said diet good quality is which FCR value is low.

Yannakopoulos & Tserveni- Gousi (1986) stated that egg weight average of quail is 10 g or 8% from the body weight. Data on table 2 showed that the egg weight was not different in all treatment, quantity linolieic acid and crude protein are influence on egg weight but in the same level of both nutrient was not significant (Kurniawan 2014). Nobakht and Safamehr (2007) stated that lysin in diet can increased egg weight of the quail. Hen day is how to calculate the daily egg production. percentage of total egg divided by total quail. The quail that fed by crickets meal has higher hen day than by indigofera meal and fish meal. This is due to the high content of protein and feed intake on the feed substituted with flour crickets. Sangilimadan *et al.* (2012) states that quails fed with protein levels of 22% have a hen day is higher than the quail fed by 19% and 16% of protein level. Cricket treatment has the higher than other treatment, but all treatment were same in egg weight. So, treatment with cricket meal can increased egg production of quail.

## Conclusion

Cricket meal and indigofera shoot leaf meal can be used as alternative protein source to substitute fish meal in quail diet. Cricket meal in ratio 8% as protein source recommended to substitute fish meal base on feed intake, protein intake, fat intake, and hen day.

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# Benefit of Kemuning Leaves Meal in Ration Containing Date Fruit Waste to Suppress Gastrointestinal Parasites Infestation of Goats

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## Abstract

*Nematodiasis is one of the obstacles in achievement optimum milk productivity. Date fruit waste (DFW) is waste from the production of date fruit juice, can be used as animal feed as an energy source, and contain flavonoid. Kemuning leaves (Murraya paniculata [L.] Jack) is a herb contain active compound as anthelmintic agent. The aim of this research was to study the influence of DFW without or with 1% kemuning leaves meal as anthelmintic agent to suppress gastrointestinal parasites (worm) infestation in PE goat. The research consists of the R0 (control with oxfendazole oral (dose 5 mg Kg<sup>-1</sup> BB<sup>-1</sup>), R1 (ration with 10% date fruit waste (DFW)), R2 (R1 with 1% kemuning leaves meal). Nine lactating Etawah crossbreed goats of 56.7±10.2 Kg body weight, number of calving of 1–4 times and average milk production 857±173 mL day<sup>-1</sup>, randomly grouped into 3 groups and each treatment 3 replicates. Data analyzed with analysis of variance (ANOVA). The parameters were feed intake, digestibility, average daily gain (ADG) and eggs per gram (EPG) of feces as indicator gastrointestinal parasites infestation. The result showed that the treatments were not influence all parameters but there was evidence that after less 6 weeks (38d and 37d) consumed the ration containing 10% without or with 1% kemuning leaves meal showed decreasing EPG similar than control. As conclusion, the ration containing 10% and with addition 1% kemuning leaves meal could be used as anthelmintic agent to solve gastrointestinal parasites infestation in PE goats.*

*Keywords: anthelmintic, date fruit waste, gastrointestinal parasites (worm), kemuning (Murraya paniculata)*

## Introduction

Population of Indonesia and awareness of the importance of animal protein were increased caused demand from livestock products increased, one of which milk, but it has not been fulfill by domestic milk productions. Domestic milk productions still low, influenced by several factors, including the number of dairy goats population low, the management of maintenance, feeding and disease prevention were less optimum. Characteristic tropical climate with high humidity caused easily livestock disease with a high prevalence of parasites infection. Parasites (worm) infection can decrease performance and productivity of dairy goats at 6.25%–21.5%, thus caused to economic losses (Alberti *et al.* 2012). The used of synthetic anthelmintic of worms has long been used, in a long-term period caused to resistance. Therefore can be required alternative anthelmintic medicines work effectively to suppress the viability of parasites, economical price and simple application.

Herbs plants contain active compounds can be used as anthelmintic, one of which kemuning. Kemuning leaves contain active compound such as tannins, coumarin, flavonoids and alkaloids that have anthelmintic activity. Kemuning leaves have 13 types of coumarin out of 39 types of coumarin in kemuning plants, 10 types of flavonoids out of 20 types of flavonoids in plants kemuning, and the type of alkaloid isyuehchuken (Ng *et al.* 2012). The content of phenol and flavonoid compounds in the methanol extract was 53 mg Kg<sup>-1</sup> and 41.92 mg Kg<sup>-1</sup> (Vagashiya *et al.* 2011). Chaira *et al.* (2009) recently reported that among the famous Tunisian dates the highest content of flavonoids was present in the Korkobbi variety (54.46 quercetin equivalents/100 g fresh weight). The results of phytochemical test qualitatively showed date fruit waste (DFW) contains many flavonoids (Yuniarti 2013). This research was aim to study the effects of kemuning leaves meal in rations based dates fruit waste as antiparasites in dairy Etawah crossbreed goats.

## Material and Methods

This experimental used 9 Etawah crossbreed goats of first to fourth lactation period, were divided into 3 similar groups and 3 feeding treatments. The average body weight were 56.7±10.2 Kg and milk

production 857±173 mL day<sup>-1</sup>. The basal ration was consisted of 35% forage and 65% concentrate. Rations have a balance of protein and energy, with the feed additives of kemuning leaves meal and 10% level dates fruit waste (DFW). Rations were based on the requirements of goats lactation, such as 12-17% crude protein and 53-66% TDN (NRC 2007). The compositions of the nutrient content of rations of this study were presented in Table 1.

Goats were housed in the individual stall barn, made of bamboo and wood with the average size of 2 x 1.5 meters. Each stall barn was equipped with a feed and a drink. Temperatures in the stall barn between 18–32.9 °C and humidity 48%–99%, 18.0–29.3 °C and 66–99% humidity in the morning and afternoon temperatures between 22.0–28.9 °C and humidity 65–99%. Feed given 3 times a day consisting of concentrates, DFW and forage. Feed given every hour 06.30, 14.00, and 16.00, and the drink was given *ad libitum*.

Feed intake was evaluated by calculating the amount of feed intake in the form of fresh rations levels multiplied by the result of the proximate analysis. Feces collection were research end, ie for 6 days to observed digestibility of DM and organic matter (OM). DM digestibility and OM was calculated as:

$$\text{Digestibility (\%)} = \frac{\text{Intake nutrients} - \text{Feces nutrients}}{\text{Intake nutrients}} \times 100 \%$$

Egg per gram (EPG) inspection carried out before granting an anthelmintic and every week during maintenance. EPG was calculated with the method of McMaster (Permin and Hansen 1998). The efficacy of each treatments were evaluated by a decrease in the number of worm eggs or faecal eggs counts reduction (FECR) with the formula as follow:

$$\text{FECR (\%)} = \frac{\text{Average EPG before treatments} - \text{average EPG after treatments}}{\text{Average EPG before treatments}} \times 100\%$$

Table 1. Composition of feed and nutrient content (% DM)

Item	Diet		
	P 0	P 1	P 2
Ingredient,% of DM			
<i>Pennisetum purpureum</i>	25.00	25.00	25.00
Pellet <i>Indigofera</i> sp.	10.00	10.00	10.00
Date fruit waste	0.00	10.00	10.00
Tempe waste	42.00	31.00	31.00
Coconut meal	15.68	16.36	16.36
Premix	0.52	0.55	0.55
White brain	5.23	5.45	5.45
<i>D icalcium phosphate</i>	0.52	0.55	0.55
CaCO <sub>3</sub>	1.05	1.09	1.09
Kemuning leaves meal *)	0.00	0.00	1.00
Nutrient composition,% of DM unless stated			
Dry matter <sup>1</sup>	43.84	44.61	44.61
Ash <sup>1</sup>	7.97	8.56	8.56
Ether extract <sup>1</sup>	5.17	5.07	5.07
Crude protein <sup>1</sup>	14.13	13.93	13.93
Crude fiber <sup>1</sup>	28.64	26.54	26.54
NFE <sup>2</sup>	44.08	46.24	46.24
TDN <sup>3</sup>	61.26	61.46	61.46

P0: control, P1: rations with 10% concentrate DFW, P2: rations with 10% concentrate DFW with kemuning leaves meal; \*) Kemuning leaves meal: 1% of the concentrate DM. <sup>1</sup> Results of laboratory analysis of Biological Resources and Biotechnology, BAU (2014), <sup>2</sup> Based on calculation (%) NFE = (%) DM [(%) Ash + (%) EE + (%) CP + (%) CF], <sup>3</sup>TDN = 37.937 - 1.018 (CF) - 4.886 (EE) + 0.173 (NFE) + 1.042 (CP) + 0.015 (CF)<sup>2</sup> - 0.058 (EE)<sup>2</sup> + 0.008 (CF) (NFE) + 0.119 (EE) (NFE) + 0.038 (EE) (CP) + 0.003 (EE)<sup>2</sup> (CP) (Hartadiet *al.* 1980).

Measurement of physiological responses of goat include rectal temperature, respiratory rate, and heart rate were analyzed twice a day with three replicates, in the morning at 6.00 to 07.30 and the afternoon at 14.30 to 16.00. FECR data were described in descriptive whereas the other data were subjected to statistical analysis by using *Analysis of Variance* (ANOVA) in a randomized complete block design.

## Results and Discussion

The effect of addition kemuning leaves meal (KLM) and date fruit waste (DFW) in the ration on feed intake and dry matter (DM) and organic matter (OM) digestibility were presented in Table 2. There were no significantly differences in the level of feed intake (DM and OM) with the addition of kemuning leaves meal in rations based date fruit waste (DFW), although there was a trend to higher feed intake in the group that without kemuning leaves meal. The percentage of dry matter intake against body weight amounted to 3.2%. NRC (2007) stated requirements DM goat lactation 2.8%–4.6%. This indicates that DFW and KLM weren't decreased palatability and the intake of dry matter and organic matter ration goats weren't disturbed by the presence of parasites (worm) infection. As well as digestibility of dry matter and organic matter were not significant differences among the treatments. Thus, it was indicated that parasites disease was not until disturbed digestibility caused by ideal feed as DFW mixture in the ration.

Anthelmintic is a chemical compound that destroy or remove worms from the gastrointestinal tract or other organs and tissues in the body of host (Permin *et al.* 1998). The impact of anthelmintic in goat presented in Table 3 and Figure 1. Calculation of the reduction of the number of eggs aimed to determine the effectiveness of used anthelmintic in goat. Oxfendazole directly reduces the number of eggs *Strongylid* sp. the fourth week was the largest decrease of 100% of before treatment due to inhibiting the activity of the gastrointestinal nematode larvae stage (Gonzalez 1997).

Table 2. Feed intake and digestibility of Etawah crossbreed goat

Parameter	Diets		
	P 0	P 1	P 2
DM intake ( g day <sup>-1</sup> )	2142 ± 430	2020 ± 280	1755 ± 295
OM intake (g day <sup>-1</sup> )	1910 ± 384	1790 ± 248	1553 ± 260
DM digestibility (%)	75.52 ± 6.30	83.21 ± 4.55	75.03 ± 4.86
OM digestibility (%)	77.88 ± 5.63	83.36 ± 4.59	76.75 ± 4.87

Reduction of the number of worm eggs kind of *Trichuris* sp. only on P2 significant drop from 700 to 60 eggs in second week treatment, then to be static until the fourth week. In other words, fecal egg count reduction (FECR) of *Trichuris* sp. -91.43%, means was decreased of 91.43% worm eggs from before treatment. Furthermore FECR *Trichuris* sp. was decreased until not found the worm eggs in the fifth week. It was a possibility due to the effected of kemuning leaves meal to suppress the development of worms, so that the worms didn't get to grow and reproduce. Thus, kemuning leaves potentially work better to suppress the number of worm eggs *Trichuris* sp. compared worms *Strongylid* sp. The result showed treatments were less 6 weeks (38d and 37d) consumed rations (P1 and P2) can be decreased similar with control (-100%). EPG factors were affected the stadium parasites, fecundity of female worms, male-female ratio of worms, immune response and experience an infection (Permin and Hansen 1998; Tizzard 1988).

Table 3. FECR *Strongylid* sp. for 5 weeks.

Week	Diets		
	P0	P1	P2
	(%)		
1	-29.72	38.42	79.87
2	-66.00	18.53	67.44
3	-70.86	18.78	69.88
4	-100.00	-18.31	-39.91
5	-100.00	-75.35	-43.67

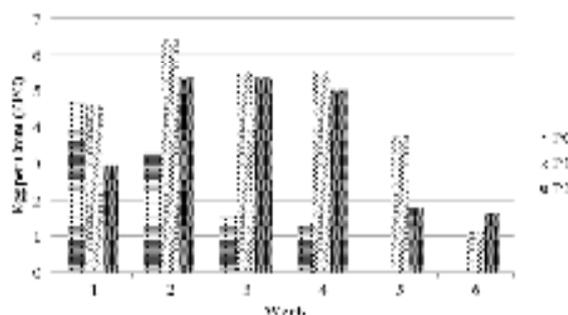


Figure 1. EPG *Strongylid* sp for 5 weeks.

Dates fruit waste not only as an energy source feed, but also has the largest active compounds flavonoids is affect against immunity, thus reduces EPG. Kemuning leaves have the most active compounds tannins, coumarin, flavonoids, alkaloids which have anthelmintic activity. The number of active compound content in the kemuning leaves and DFW allowed mechanism of synergistic or antagonistic, so it could not be predicted compounds that can inhibit or enhance these herbs work activities.

Tannins inhibit egg hatching and infective larvae development of worm by reducing the availability of nutrients for the growth of infective larvae, disrupting oxidative phosphorylation reaction or binding of free proteins in the digestive tract of the host body or glycoprotein on the cuticle of parasite (Gulnaz and Salvitha 2013; Min and Hart 2003). These activities can interfere with the metabolism and homeostasis in the body of the worm and the worm will eventually die due to lack of ATP. In addition, the collagen matrix with reactive tannins caused the loss of flexibility, hence the worm didn't move and non-functional causes paralysis followed by death (Gulnaz and Savitha 2013).

Physiological responses of Etawah crossbreed goats belong to normal such as heart rate 81.11–102.11 times  $\text{min}^{-1}$ , respiratory rate 27.27–52.99 times  $\text{min}^{-1}$  and a rectal temperature of 38.62 °C–39.95 °C. Qiston and Suharti (2005) reported that the physiological response of goats were 86.6 times the heart rate  $\text{min}^{-1}$ , respiratory rate 67.6 times  $\text{min}^{-1}$  and rectal temperature of 38.7 °C. The results showed that the physiological response P1 and P2 with controls (P0) were not significantly different. Thus the rations didn't affect on the physiological response of goats.

Table 4. Physiological response of Etawah Crossbreed Goats

Parameter	Time	Diets		
		P 0	P 1	P 2
Heart rate (times $\text{min}^{-1}$ )	Morning	81.11 ± 8.68	89.96 ± 11.03	87.54 ± 14.16
	Afternoon	92.99 ± 10.67	91.38 ± 8.10	102.11 ± 15.65
Respiratory rate (times $\text{min}^{-1}$ )	Morning	27.27 ± 6.99	37.22 ± 10.77	32.23 ± 10.25
	Afternoon	41.22 ± 12.14	57.08 ± 16.40	52.99 ± 11.49
Rectal temperature (°C)	Morning	38.62 ± 0.20	38.66 ± 0.18	38.67 ± 0.18
	Afternoon	39.01 ± 0.18	38.95 ± 0.22	39.11 ± 0.19

## Conclusion

It is concluded that combinations of date fruit waste and kemuning leaves meal were potential as anthelmintic to reduce parasitic (worm) egg counts *Strongylid* sp. and *Trichuris* sp. and could be an alternative solution as anthelmintic, to lower the risk of disturbance of the parasite on goat, but with slow activities.

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# Golden Snail Eggs (*Pomacea canaliculata*) and Bay Leaf Meal as Natural Feed Supplement to Improve Quail Egg Quality and Reduced Yolk Cholesterol

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## Abstract

Golden snail eggs have calcium and carotenoid content that is useful as an antioxidant and a natural feed supplement. Bay leaf contain essential oils, eugenol, flavonoids, saponins, tannins that are useful as natural antioxidants and can be used to reduce cholesterol content in egg yolk. This study aimed to evaluate the effect of golden snail eggs and bay leaf to improve the quail egg quality and to reduce cholesterol content in quail egg yolk. The material used were 120 pullet quail, golden snail eggs, bay leaf meal, laying quail commercial feed. A completely randomized design (CRD) with ANOVA and Duncan used in this study. There were four treatments and three replications with 10 quails each treatment. The treatments were control diet without addition of golden snail eggs and bay leaf meal (P0), and diet with addition of golden snail eggs and bay leaf meal as much as 1% & 0% (P1), 0% & 1% (P2), and 1% & 1% (P3) of the control diet. Parameters observed were egg weight, Haugh units (HU), and cholesterol content of quail egg yolk. The addition of golden snail eggs (P1) was significantly produced heavier egg weight than other treatments. The addition of 1% golden snail eggs and 1% bay leaf meal (P3) was significantly increased Haugh Unit and decreased cholesterol content of the yolk quail as much 8.57%. Golden snail eggs and bay leaf meal can be use as natural feed supplement in laying quail diet to produce better egg quality and reduce yolk cholesterol.

Keywords: bay leaf meal, cholesterol, golden snail eggs, quail egg quality

## Introduction

Snails (*Pomacea canaliculata*) is one type of freshwater snails that are easily found in rice fields or lakes. Snails is one cause of damage to agricultural crops in Indonesia (Budiyono, 2006). Apart from being a pest, golden snail eggs have benefits in the world of farming although until now its use has not been optimally. Golden snail eggs have a macro-micro minerals and carotenoids quite high, such as calcium (Ca), magnesium (Mg), potassium (K), sodium (Na), iron (Fe) and zinc (Zn). The content of carotenoids in eggs snails higher than carrots (Ameliawati 2013). Cholesterol and the mineral content of macro-micro high can improve the quality of quail eggs. Bay leaves (*Syzigium polyanthum*) is a spice commonly used in Indonesia. Bay leaves contain essential oils with essential components eugenol, flavonoids, tannins, and saponins which can be used as an antioxidant and can cause lower cholesterol levels in quail eggs (Saputra 2008). Giving flour golden snail eggs and flour bay leaves as a feed supplement in the ration is expected to improve the quality of eggs and lowering cholesterol levels in quail. This study aimed to evaluate the utilization of golden snail eggs and flour bay leaf on the quality and cholesterol quail egg.

## Materials and Methods

This research was conducted at the Laboratory cage C and Poultry Nutrition Laboratory Level 3 Faculty of Animal Husbandry, Bogor Agricultural University in March-May 2015. Livestock used are quail ready for production (age 4-5 weeks) of 120 birds. Materials used in this study are bay leaves, golden snail eggs, quail rations, rice flour, water, detergent, disinfectant, and lime. The tools used are 60°C oven, oven 105°C, digital scales, thermometers, paper cement, stainless steel container, blender, egg tray, gauges yellow egg or yolk color fan (Roche), hygiene kits, glass, stationery, caliper, petri dish, where food and drinking water.

Bay leaves cleaned and dried in an oven at 60°C (dry sun) for 24 hours at a dose of 1 kg per one drying. The last stage, bay leaf grinded to form a blended manner. Golden snail eggs dried in an oven at 60°C (dry sun) for 24 hours. Once dry, the egg-shaped snails crushed to powder with the addition of rice flour as a filler. Maintenance quail will be maintained during the 45-day period in which the 24-day and 21-day adaptation period giving golden snail eggs, flour and flour bay leaf. Feed given as much as 24 grams / head /

day, while the drinking water provided ad libitum. Quality chemical egg which include cholesterol content of the yolk, protein, fat, carbohydrates, and minerals. Cholesterol content analysis measured by the method of Liebermann Burchard (1885).

This study used a completely randomized design (CRD), with 4 treatments and 3 replications with each replication using 12 quails. Ration treatment to be given are:

P0 = control ration without flour golden snail eggs and flour bay leaf

P1 = ration with the addition of 1% flour golden snail eggs and flour bay leaf 0%

P2 = ration with the addition of flour 0% golden apple snail eggs and 1% flour bay leaf

P3 = ration with the addition of 1% flour golden snail eggs and 1% flour bay leaf.

Data obtained from this study was analyzed using ANOVA (Analysis of Variance or ANOVA). the different of mean treatments was tested further in the form of Duncan test. The parameters measured were the average consumption, feed conversion, the average egg mass production, the average weight of the egg, quail egg production, Haugh unit, yolk color quail egg, egg yolk cholesterol content, and nutrient content of quail eggs.

## Result and Discussion

### Chemical Composition Golden Egg Snail (*Pomacea canaliculata*)

One way to determine the chemical composition of the golden snail eggs by performing the proximate analysis in order to determine the nutrient content, which includes ash, protein, fat and crude fiber.

Table 1. Data analysis proximate composition flour golden snail eggs with literature data proximate analysis golden snail eggs

Chemical Composition	Golden egg snail	
	Proximate analysis <sup>1</sup>	literature of proximate analysis <sup>2</sup>
Crude protein	7.24	3.32
Ash	7.525	13.81
Crude Fat	0.26	0.19
Crude fiber	0.115	-

Source:

<sup>1</sup> Laboratory PAU, Bogor Agricultural University; <sup>2</sup> Pambudi (2011).

Based on Table 1. there is a difference between the proximate analysis data with literature data the analysis of proximate, this happens because the materials used to make the proximate analysis are adding filler with rice flour. Improvement and deterioration occur in each nutrient levels. On crude protein and crude fat increased while the ash decline, this happens because as comparison of flour more than the golden snail eggs.

### The Average Consumption And Conversion Ration (FCR)

Based on Table 2. the average consumption of feed rations with P3 treatment was higher compared with other treatments, while treatment P0 has the lowest average consumption compared with other treatments. Feed conversion ration treatment P3 has higher FCR while the P0 have low FCR. Conversion rate over time might indicate that the feed used more efficiently. Research Achmanu *et al.* (2011) showed quail feed conversion was 2.45. The high value of FCR in his research-resulting from the additional treatment was due reached peak production age yet.

### Mean Production of Eggs Mass

The average egg production mass showed. In Table 2, treatment P0 has egg production mass higher than treatment P3. This happens because the P3 treatment with the addition of bay leaves and golden snail egg. The high crude fibre content in the ration caused fast motility of digestive tract and decreased of nutrient absorption. Fermented products such short-chain fatty acids which are mainly acetate, propionate, and butyrate (Jacobasch *et al.*, 1999).

### The Average Weight of Eggs

Weight of eggs is a qualitative trait that can be derived. Factors that may affect to the weight of the egg is a type of feed, the amount of feed, genetic, environmental enclosures, egg-laying period, and the size

of the parent body (Yuwanta, 2004). Important factors that greatly affect to the egg weight is protein and amino acids content in the diet and the amount of linoleic acid (Wahju, 1982). Table 2 shows that the quail eggs produced by treatment with the addition of 1% flour golden snail eggs have the highest average of egg weight ( $10.510 \pm 0.430$  g) among other treatments, while quail eggs produced from treatment 1% addition of bay leaf flour alone has the average egg weight lower ( $9.765 \pm 0.423$  g). Egg weight variation caused by the amount of nutrients in the feed of different treatments. In P3 (1% flour golden snail eggs and 1% flour bay leaf) has a complementary effect. Saponins at bay leaf flour would inhibit the synthesis of fat while flour golden snail eggs as a protein will improve the egg weight.

Table 2. Feed intake, FCR and egg quality of quail

Treatment	P0	P1	P2	P3
Feed consumption (g/head/day)	$23.23 \pm 0.0619$	$23.66 \pm 0.3396$	$23.87 \pm 0.0204$	$24.12 \pm 0.0035$
Conversion ration (FCR)	2.690 <sup>a</sup>	3.021 <sup>ab</sup>	3.216 <sup>ab</sup>	3.334 <sup>b</sup>
egg mass (g/head)	$8.359 \pm 0.542^a$	$8.102 \pm 0.309^a$	$8.250 \pm 1.187^a$	$7.237 \pm 0.515^a$
Egg Weight (g)	$10.062 \pm 0.525^a$	$10.510 \pm 0.430^b$	$9.765 \pm 0.423^a$	$9.896 \pm 0.392^{ab}$
Total egg weight (g)	$539 \pm 2.100^a$	$467 \pm 2.247^{ab}$	$478 \pm 2.174^{ab}$	$460 \pm 2.060^b$
Haugh Unit	$92.20 \pm 5.94^b$	$84.01 \pm 4.79^a$	$86.80 \pm 8.84^{ab}$	$91.13 \pm 6.16^b$
Cholesterol quail egg yolk (mg/ 100g)	$485.50 \pm 17.75$	$472.16 \pm 101.63$	$448.44 \pm 19.63$	$443.87 \pm 67.28$

Different superscript in the same line means significantly different ( $P < 0.05$ )

P0 = control ration without flour golden snail eggs and flour bay leaf

P1 = ration with the addition of 1% flour golden snail eggs and flour bay leaf 0%

P2 = ration with the addition of flour 0% golden apple snail eggs and 1% flour bay leaf

P3 = ration with the addition of 1% flour golden snail eggs and 1% flour bay leaf.

## Egg Production

Egg production has a close connection with the consumption and nutrient content of feed. High protein content with the adequacy of energy and calcium more influence on egg production (Cheeke, 2005). Quail with treatment control diet had the highest egg production is 53.472%, while the quail with the addition of 1% of flour treatment golden snail eggs and flour bay leaf 1% have the lowest (45.63%) (Table 2). This is caused by the function of the saponin as a substance that inhibits the absorption of protein. These results are still considered not good enough because the quail with the age of 10-20 weeks were given the protein content of 20-22% with the lighting 22 hours / day have ranged from 63 to 71.7% egg production (Eishu, 2005).

## Haugh Unit

Haugh unit is used as a quality parameter calculated based on the freshness of high egg whites and egg weight (Siregar, 1994). The addition of the feed treatment in this study have not affect to the process of formation of albumen. The results obtained was similar with Kurnia *et al* (2012) that the quail egg Haugh unit is 95.63. Wahju (1988) argue that methionine is the first limiting amino acid or amino acid critical first albumen structures often affects the formation and stabilization meshes affect ovomusin. Thus, the fulfillment of methionine, the more stable formation of ovomusin.

## Cholesterol content of Quail Egg Yolk

The cholesterol content was high when compared with the cholesterol content of the yolk according to research results Latif *et al* (2011) that the quail egg yolk cholesterol was 202.00 mg / dl at age 4 weeks of laying quail. The addition of 1% flour golden snail eggs and 1% flour bay leaf proven to reduce cholesterol levels highest in this study as much as 8,57%. Saponins in flour bay leaf and carotenoid content in flour golden snail eggs cooperate with each other in reducing cholesterol levels in the quail eggs. Supplementation materials containing carotenoids for example flour golden snail eggs in poultry rations can produce low-cholesterol eggs. The ability of carotenoids in lowering cholesterol in two ways: 1)  $\beta$ -carotene are antioxidants that can prevent lipid oxidation, and 2)  $\beta$ -carotene is able to inhibit the action of the enzyme HMG-CoA reductase activity so as not to form mevalonate required for cholesterol synthesis (Einsenbrand 2005 and Sies and Stahl, 1995). Saponins proven to reduce the synthesis of fatty acids (Santoso 2010).

## Egg Yolk color

The color of the yolk is strongly influenced by the feed. Feed containing a high carotenoid will enhance yolk color. In poultry rations is the source of carotene including corn. Carotenoids are a group of colored pigments of yellow, orange, red-orange, as well as soluble in oil (lipids). In addition, the carotenoids also a provitamin A compound (Winarno 2008). Cheesman (1958) suggested that the pigment carotene in golden snail eggs can be separated with alcohol, acetone, and pyridine, and be absorbed by the aluminum hydroxide gel. In the provision of treatment of feed, with treatment P1 and P3 with the addition of golden snail eggs can increase egg yolk color compared to the feed treatment P0 and P2 without the addition of golden snail eggs (Figure 1). This is because the golden snail eggs are high carotenoid content. Total carotenoids golden snail eggs are  $313.48 \pm 19.73$  ppm. Golden snail eggs carotenoid content of about 72 nmol / gram. The main carotene in golden snail eggs is astaxanthin. Astaxanthin is a powerful antioxidant. Golden snail eggs containing astaxanthin consisting of free form (40%), the form of monoester (24%), and forms diester (35%) esterified with fatty acids 16: 0. High carotene content in the golden snail eggs expected to be utilized as a source of the pigment that gives color to the quail eggs. In addition there is a positive correlation between the quality of the females with the content of carotenoids in eggs, so the higher the content of carotenoids in egg then the better the quality. Female parent body tissues and the concentration of carotenoid in egg reflects dietary intake of carotenoids by the mother (Garner *et al.* 2010).

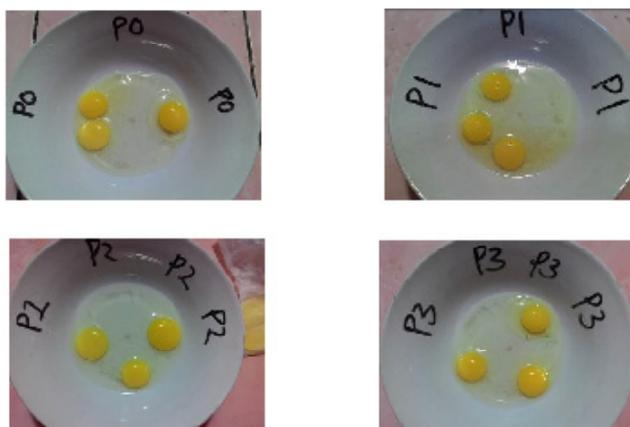


Figure 1. Quail eggs control treatment (P0), flour golden snail eggs (P1), bay leaf flour (P2), and the golden snail eggs, flour and bay leaves (P3)

## Conclusion

Quail response to supplementation of flour golden snail eggs and flour bay leaf in feed is palatable. Giving 1% flour golden snail eggs and 1% flour bay leaf caused lowers cholesterol yolk as much as 8:57%. Giving 1% flour golden snail eggs, quail eggs increase the weight significantly and increase egg yolk color becomes more reddish. Value Haugh unit (HU) in the provision of 1% flour bay leaf increased significantly.

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# ***In Vitro* Study of Calcium Soap-Soybean Oil Addition in The Rumen of Bali Cattle on Rumen Microbial Population, Microbial Protein Synthesis, Cellulase Activity, and Nutrient Digestibility**

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## **Abstract**

*Supplementation of unsaturated fatty acid from vegetable oils, such as soybean oil, in the beef cattle ration, have potency to improve the production and meat quality. However, the use of vegetable oils as sources of unsaturated fatty acids, need to be protected using calcium soap method to avoid biohydrogenation by rumen bacteria which convert the oil into saturated fatty acid. The experiment was aimed at evaluating the effect of soybean oil addition with and without protection using calcium soap in the diet on nutrient digestibility and rumen microbe in vitro. This study used randomized block design with 3 treatments and 4 replications. The treatments were control diet (C), C + 5% soybean oil (M), and C + Calcium soap of soybean oil 5% (S). The variables measured were pH value, NH<sub>3</sub> concentration, dry matter digestibility (DMD), organic matter digestibility (OMD), crude protein digestibility (CPD), ether extract digestibility (EED), crude fiber digestibility (CFD) this can be shorter becoming dry matter and nutrient digestibilities, populations of bacteria and protozoa, microbial protein synthesis and cellulose enzyme activity. The result showed that the use of soybean oil at level 5% either with or without protection using calcium soap did not affect pH value, NH<sub>3</sub> concentration, dry matter and organic matter digestibility, crude protein digestibility, crude fiber digestibility and ether extract digestibility. Population of protozoa also similar among treatments, but the addition of Ca-soap soybean oil tend to increase bacteria population. Supplementation of Ca-soap reduced rumen microbial protein synthesis, meanwhile it could increase the activity of cellulase enzymes. This result indicates that the addition of soybean oil with or without protection using calcium soap at level 5% in the concentrate ration have positive effect on bacterial population and cellulase enzymes activity, but it did not alter feed digestibility.*

**Keywords:** *bali cattle, calcium soap, nutrient digestibility, soybean oil*

## **Introduction**

The increase in Bali cattle productivity should be followed by improving meat quality, particularly unsaturated fatty acid content. Naturally, meat products from ruminants have high content of saturated fatty acid because of bio-hydrogenation process by rumen microbe. Rumen bacteria convert unsaturated fatty acid from feed to become saturated fatty acid and subsequently increase saturated fatty acids which are absorbed by the animals.

One of strategy to increase the poly unsaturated fatty acid (PUFA) content of ruminant meat is supplementation with PUFA sources from plant oil such as soybean oil. Soybean oil is rich in PUFA mostly linolenic acid 15-64%, oleic acid 11-60%, linoleic acid 1-12% and arachidonic acid 1.5% (Muliawati 2006). Soybean oil as PUFA sources in the ruminant diets have to be protected to avoid bio-hydrogenation processes by rumen bacteria. Moreover, the use of fat sources in the high level has disadvantage effect on rumen degradation and fermentation. Previous research showed some protection technology to shield PUFA sources from bio-hydrogenation process is coating using formaldehyde, alkali, calcium soap or microencapsulation. The cheap and applicable protection method is using calcium soap (Jenkins and Palmquist 1984).

This research was aimed at evaluating the effect of Calcium Soap Soybean oil (Ca-soap SO) addition at a level of 5% in the ration on *in vitro* nutrient digestibility, rumen microbial populations, microbial protein synthesis and cellulase activity using rumen fluid of Bali cattle.

## Materials and methods

### *In vitro* fermentation

The soybean oil were obtained from CV. MH. Farm Bogor. Calcium soap from soybean oils was produced according to Kumar *et al.* (2006). *in vitro* fermentation was conducted according to Tilley and Terry method (1963). The rumen fluid for this experiment was obtained before morning feeding from the rumen of fistulated Ongole crossbred beef cattle consuming commercial concentrate and elephant grass. The substrate contained 60% king grass forage and 40% concentrate mixture (cassava by-product, wheat pollard, soybean meal, coconut cake meal, molasses, CaCO<sub>3</sub>, premix, urea, and oil) with 15-17 %CP and 69-74% TDN (Table 1 and Table 2). Samples for counting protozoal and bacterial population, microbial protein synthesis and cellulose enzyme activity was taken at 4 h incubation. Dry matter and organic matter digestibilities were evaluated after 48 h incubation.

### Sample Analysis

Protozoal numbers were counted under a microscope, and bacterial population were determined using roller tube method (Ogimoto and Imai 1981). Microbial protein synthesis was analyzed according to the method of Makkaret *al.* (1982) and Lowry *et al.* (1951). Cellulase enzyme activity was analysed using method described by Patra *et al.* (2006).

Table 1. Concentrate composition on dry matter basis

Ingredient	Use (% DM)		
	Control (C)	C + soybean oil (SO)	C + calcium soap soybean oil (Ca-soap SO)
Cassava waste	30	25	25
Pollard	31.5	31.5	31.5
Coconutcake meal	20.5	20.5	20.5
Molasses	15	15	15
CaCO <sub>3</sub>	1.5	1.5	1.5
Urea	1.5	1.5	1.5
Soybean oil	0	5	0
Calcium soap soybean oil	0	0	5

Table 2. Chemical composition of experimental substrate on dry matter basis with 60% king grass forage and 40% concentrate mixture

Treatments	DM	Ash	CP	EE	CF	NFE	TDN*	Ca	P
C	87.12	6.65	13.15	3.83	17.87	59.29	68.87	0.70	0.31
SO	84.57	6.55	13.08	6.82	17.56	56.77	66.70	0.70	0.31
Ca-soap SO	87.26	7.13	13.08	6.18	17.56	56.77	66.70	0.70	0.31

DM (Dry Matter), CP (Crude protein), EE (Ether Extract), CF (Crude Fiber), NFE (Nitrogen Free Extract), TDN (*Total Digestibility Nutrient*), Ca (Calcium), P (Phosphor).

\*TDN= 70.6 + 0.259 x CP + 1.01 x EE – 0.76 x CF + 0.0991 x NFE (Hartadi *et al.* 1980)

### Experimental Design

This study used randomized block design with 3 treatments and 4 replications. The treatments were control diet (C), C + 5% soybean oil (SO), and C + calcium soap of soybean oil 5% (Ca-soap SO). The variables measured were pH value, NH<sub>3</sub> concentration, populations of bacteria and protozoa, microbial protein synthesis, cellulose enzyme activity, total gas and methane productions, dry matter digestibility (DMD), organic matter digestibility (OMD), crude protein digestibility (CPD), ether extract digestibility (EED), crude fiber digestibility (CFD).

### Statistical Analysis

All data were analyzed by ANOVA using the GLM procedures (SPSS 13.0 for windows, 2004). The significant differences among treatments were determined by Duncan's multiple range test.

## Results and Discussions

The addition of soybean oil or Ca-soap soybean oil at a level of 5% in concentrate did not affect pH value, NH<sub>3</sub> concentration, nutrient digestibility, rumen microbial population, total gas and methane productions, and microbial protein synthesis. The use of Ca-soap soybean oil tended to increase cellulose enzyme activity (Table 3).

The rumen pH values were still in the normal condition which was in the range of 6.78-6.82. This result indicates that the addition of soybean oil or Ca-soap soybean oil did not alter rumen microbial activity. Supplementation of soybean oil or Ca-soap soybean oil did not affect nutrient digestibility. This might be due to the low level of soybean oil added on concentrate.

Table 3. Effect of soybean oil addition on *in vitro* fermentation characteristic, nutrient digestibility, rumen microbial population, microbial protein synthesis and cellulose enzyme activity

Variables	Treatment		
	Control (C)	C + SO	C + Ca-soap SO
pH	6.78±2.60	6.78±2.60	6.82±2.61
NH <sub>3</sub> (mM)	10.53±2.08	9.55±2.14	9.73±2.33
Nutrient digestibility (%)			
Dry matter	73.52±8.57	71.33±8.45	72.62±8.52
Organic matter	73.33±8.56	71.25±8.44	72.50±8.51
Crude protein	74.78±8.65	73.56±8.58	75.22±8.67
Crude Fiber	80.70±8.98	79.08±8.89	80.09±8.95
Ether extract	46.90±6.85	51.57±7.18	48.85±6.99
Protozoal population (x 10 <sup>5</sup> cell/mL)	3.0±0.13	2.7±0.14	3.1±0.18
Bacterial Population (x 10 <sup>8</sup> cell/mL)	1.9±0.15	2.9±0.20	6.6±0.73
Total gas production (mL/200 mg DM)	47.44±3.71	44.61±3.95	42.04±4.02
Total gas production (mL/200 mg DMdig.)	324.10	314.68	289.64
Methane production (% of total gas)	17.90±11.39	19.52±9.24	18.55±7.30
MPS (mg/10ml)	28.32±9.77	27.78±8.48	26.05±5.99
MPS Efficiency	13.50±4.66	12.87±3.93	12.27±2.82
Cellulase Enzyme Activity	164±89.69	114.56±29.61	229.50±122.66

Supplementation of Ca-soap soybean oil slightly increased protozoal and bacterial population compared to the control treatment indicating that soybean oil protected with calcium soap technology could stimulate the growth of rumen microbes. This might be due to the calcium content of vegetable oil-calcium soap which was important for the growth of microbial cell wall. Previous research showed that oil supplementation without protection at a level of 5.0% decreased protozoal population significantly (Sitoresmi *et al.* 2009). Similar results had been reported by Adawiyah (2007); supplementation with non-protected fish oil at 1.5% had significantly decreased total bacterial population, but protozoal population was not reduced by supplementation with calcium soap protected fish oil at a level of 3%.

Total gas production decreased slightly with the addition of Ca-soap soybean oil compared to the control treatment. This result indicates that addition of Ca-soap soybean oil could improve rumen fermentation efficiency reducing total gas production. The proportion of methane production increased slightly with the addition of Ca-soap soybean oil compared to the addition of unprotected soybean oil. Microbial protein synthesis also tended to decrease with the addition of Ca-soap soybean oil, but the cellulose enzyme activity tended to increase.

## Conclusion

The additions of soybean oil with or without protection using calcium soap at a level of 5% in the concentrate have positive effect on bacterial population and cellulose enzyme activity, and did not alter feed digestibility and fermentation.

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# Incremental Level of *Chromolaena Odorata* in Complete Diet Does not Impair Intake, Rumen Fermentation and Microbial Protein Synthesis Efficiency in Cattle

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## Abstract

An emerging concern over the use of *Chromolaena odorata* as an alternative protein source for livestock is the presence of various anti-nutrient compounds in the plant's tissues. However, recent studies have revealed that physical treatments such as pelleting can effectively eliminate the anti-nutrition associated effects. This experiment aimed at quantifying effects of different levels of *C. odorata* in a complete diet (given in the form of mash) on nutrient intake, digestibility, concentration of volatile fatty acids (VFA), and efficiency of rumen microbial crude protein synthesis (EMP). Four growing Bali bulls (aged  $\pm$  2 y.o) were allotted into four dietary treatments following Latin Square Experimental Design principles. The treatments were a complete diet containing 10% (COM10) or 20% (COM20) or 30% (COP30) or 40% (COM40) of *C. odorata* meal given at 2% liveweight. The basal diet (*Sorghum plumosum* hay) and drinking water were given *ad libitum*. The tested diets were designed to be iso-protein (18%) and metabolisable energy (12 MJ) to support a minimum liveweight of 0.8 kg/head/day. Results indicated that increasing level of *C. odorata* from 10% to 40% in the complete diet did not significantly impair all variables measured, though at the highest level (40%) of inclusion, all variables have shown a diminishing trend. It can be concluded that *C.odorata* can potentially be utilized as a cheap protein source for fattened cattle when provided up to 40% in the total diet, but might have a negative effect when the level is raised above the current level.

**Keywords:** cattle, *Chromolaena odorata*, intake, microbial crude protein, rumen digestion

## Introduction

Despite its negative effect on pasture production in the dryland areas of the world (McFayden, 2004; Prawiradiputra 2007), *Chromolaena odorata* is a potential feed source due to its high crude protein content (21%) and biomass production (Mullik 2002). However, its usage as feed is hampered by the presence of various anti-nutrient compounds in the plant tissues (Aro *et al.* 2009). Therefore strategies are needed to eliminate these anti-nutrient compounds. A recent study (Mullik *et al.* 2014) revealed that physical treatments such as pelleting can effectively eliminate the anti-nutrition associated effects. The present experiment aimed at assessing the efficacy of *C. odorata* meal inclusion in a complete diet that given in the form of mash to cattle at a gradual level on intake and rumen fermentation.

## Materials and Methods

Four young male Bali cattle with an initial liveweight of 143kg ( $\pm$ 7.11 kg) were allotted into a 4 x 4 Latin Square experimental design to test four diets. The diets were complete diet containing 10% (COM10) or 20% (COM20) or 30% (COP30) or 40% (COM40) of *C. odorata* meal given at 2% liveweight. Feed allowance was adjusted for each period based on the liveweight at recorded at the end of each treatment period. Basal diet (*Sorghum plumosum* hay) and drinking water were given *ad libitum*. The tested diets were designed to be iso-protein (18%) and metabolisable energy (12 MJ) across all treatments to support a minimum liveweight gain of 0.8 kg/head/day. Each treatment period lasted for 15 days (10 days adaptation to the new diet and 5 days data collection).

Feed intake, digestibility, and rumen fermentation were measured for 5 days in each treatment period. Concentration ammonia and volatile fatty acids in the rumen fluid were measured by sampling rumen fluid (using stomach tube aspiration under vacuum) three hours after feeding on the last day of each treatment period. The rumen liquor was strained through two layers nylon stocking and then acidified with

concentrated sulphuric acid to lower the pH below 3. Molar proportion of volatile fatty acids (VFA) was quantified using gas liquid chromatography (HP 8530A with a HP 18850A GC terminal and HP 3396A integrator).

To measure rumen microbial protein production (MCP), spot samples of voided urine by each cattle were taken daily during each data collection period. Daily spot samples were bulked for each animal in a glass container. The collected urine was acidified using 10% H<sub>2</sub>SO<sub>4</sub> to keep the pH below three. At the end of each data collection period, the bulk urine samples were processed and analysed following standard protocols for spectrophotometry proposed by Chen and Gomez (1995). Calculation of MCP and efficiency of MCP was performed using the formula for Zebu cattle proposed by (IAEA 2003). The MCP was calculated using formula  $Y = 0.85 X + 0.147 W^{0.75}$ , where Y is total purine derivatives (PDs) excreted in the urine; 0.85 is proportional recovery of absorbed purines in urine; X is total microbial purines absorbed; 0.147 is coefficient for endogenous purine derivatives in the urine for *Bos indicus* cattle, and  $W^{0.75}$  is metabolic weight of cattle. Since spot urine samples were used, molar ratio of PDs:creatinine was used to estimate daily excretion of PDs. Daily creatinine excretion was assumed to be constant and a value of 0.91 mmol/kg W<sup>0.75</sup> (Chen *et al.* 1996) was adopted in the computation.

Data were statistically analysed using General Linear Model principles (univariate) for Latin Square Experimental Design. Differences between treatments were detected at  $P \leq 0.05$  using SAS statistics software.

## Results and Discussion

### Intake and Digestibility

All parameters of intake and digestibility were not significantly affected by inclusion of chromolaena meal in the diet, though the empirical data showed a diminishing trend as the level of chromolaena meal increased (Table 1). Total dry matter intake ranged from 2.66 to 3.01% LW (Table 1). Lack of statistical difference in the present study most probably due to a large within treatment variation (larger SEM value) compared to those of pelleted diets. The larger variability in intake of mash diet mainly related to physical form of the diet since some animal could be distracted by dust from the mash as it can block the breathing airways. Other intake parameters (organic matter and crude protein) were also following the DMI trend.

Ratio of crude protein intake (CPI) and digestible organic matter intake (DOMI) is a good nitrogen-carbon balance indicator for rumen function and efficiency of nutrients utilization at tissue level. Figures in the present experiment were in the range of 224-256 g protein/kg DOMI (Table 1). With 75% digestibility of crude protein (Table 1), it should be estimated that the ratio of digestible protein-DOMI was at least 170 g DCP/kg DOMI which was a very good balance for nutrient fermentation in the rumen and utilization at the tissue.

Table 1. Intake and *in vivo* digestibility of a complete diet contains *Chromolaena odorata* meal at a rate of 10% (COM10) or 20% (COM20) or 30% (COM30) or 40% (COM40) and provided to cattle in mash form

Variable	COM10	COM20	COM30	COM40	SEM	P value
<i>Total intake</i>						
Dry matter (kg <sup>h</sup> )	4.16	3.98	3.56	3.58	0.028	0.105
Dry matter (% liveweight)	3.01	2.91	2.66	2.72	0.018	0.017
Organic matter (kg <sup>d</sup> )	3.78	3.57	3.10	3.19	0.024	0.061
Crude protein (g <sup>d</sup> )	558	547	477	450	0.156	0.060
Digestible organic matter (kg <sup>d</sup> )	2.29	2.21	1.98	2.16	0.011	0.296
Protein:digestible organic matter intake	224	222	256	232	0.125	0.711
<i>Digestibility</i>						
Dry matter (%)	61.6	62.4	56.0	57.3	8.201	0.307
Organic matter (%)	68.0	64.9	62.1	62.7	6.950	0.441

Insignificant decline in the intake found in the present experiment is similar to the results reported by Mullik *et al.* (2014) for cattle given pelleted diet that had the same nutrient composition and the same level of chromolaena meal or given to sheep (Apori *et al.* 2001). This trend could not be related to nutrition composition aspect since diets for all treatments were formulated to provide the same level of energy (12

MJ/kg DM) and protein (18%). Therefore, the most probable factor suppressing the intake was secondary metabolic compounds in the diet as in chromolaenameal. Heat drying and grinding of chromolaenamight fail to eliminate anti-nutrient compounds in the meal. A recent study in Institut Pertanian Bogor (Y.M. Mulik, unpublished data) has found that total tannine and anti-trypsin concentration in chromolaena leaf increased dramatically to 3 fold when oven dried or sun-dried. This might be the underlying explanation for the negative effect in intake since it is well documented that anti-nutrient compounds upset rumen fermentation and nutrient digestibility leads to a negative feedback on the intake.

Eventhough digestibility variables were not affected significantly when the level of chromolaena meal was increased up to 40%, yet there was a decline by up to 10% for DM digestibility (DMD) and 9% for organic matter digestibility (OMD). However, digestibility coefficient found in the present experiment (Table 1) was reasonably good since there is a good balance of nutrients and high level of mash intake (up to 2% LW).

### Rumen Fermentation and Microbial Crude Protein Production

Measurement of rumen fermentation products such as pH, ammonia (NH<sub>3</sub>), volatile fatty acids (VFAs), microbial protein (MCP) production and efficiency of synthesis (E<sub>MCP</sub>) showed insignificant effects (Table 2). Rumen fluid pH tended to shift slightly toward base zone (7.02-7.14). This happens because of the high protein content (18%) in the diet. Rumen NH<sub>3</sub> was in the range of 262-268 mg/L which indicates an adequate supply of nitrogen for an optimal rumen function. This high rumen NH<sub>3</sub> partly arises from the urea used the diet to adjust the protein level to 18%. Total VFAs and molar propotion of these rumen fermentation products were also not affected by the level of chromolaena meal. Insignificant effects of incremental level of chromolaena in pellet diet on these variables was also reported by Mullik *et al.*(2014) for cattle. Again, lack of response to levels of chromolaenameal was expected as the same composition of diet presented to the rumen of cattle in all treatemets.

Table 2. Rumen fermentation and microbial crude protein (MCP) production in cattle given a complete diet contained 10%(COM10) or 20%(COM20) or 30%(M30) or 40%(COM40) *Chromolaenaodorata* meal and provided in mash form

Variable	COM10	COM20	COM30	COM40	SEM	P value
Rumen pH	7.02	7.17	7.03	7.14	0.009	0.512
Rumen NH <sub>3</sub> -N (mg/L)	262	263	264	268	135.0	0.983
Total VFA (mM)	151.2	147.7	140.5	149.5	69.04	0.809
Acetat (mM)	87.2	92.4	81.7	87.2	40.05	0.713
Butirat (mM)	20.5	17.4	20.9	16.4	3.210	0.222
Propionat (mM)	43.4	45.2	37.9	44.3	0.326	0.358
Rumen MCP:						
Production (g/d)	210	232	194	172	248.2	0.163
Efficiency (g MCP/kg DOMI)	83	100	103	90	41.15	0.245

Optimal efficiency of rumen microbial crude protein synthesis (E<sub>MCP</sub>) recomanded in the current feedings standards (SCA 2007; NRC 2000) is around 130 g MCP/kg DOMI. This equal to 130 rumen digestible protein (RDP) per each kg DOMI. The RDP was not measured in the present study hence the RDP:DOMI ratio could not be established. However, the healthiness of the rumen function could be detected using E<sub>MCP</sub>. The E<sub>MCP</sub> presented in Table 2 showed a range value of 83-103 g MCP/kg DOMI. These values were far below the recomanded level. Using this data, one can confidently stated that the diet used in the present study did not support an optimum rumen microbial production. A much lower E<sub>MCP</sub> values (60.4-74.3 g MCP/kg DOMI) were also documented by Mullik *et al.*(2014) when complete diets (pellet) contained the same nutrient composition and levels of chromolaena meal were offered to cattle in a parallel experiment. However, this does not imply that the diets used in the present study did not provide adequate nutrients into the rumen since it contained 18% CP and 12 MJ ME/kg DM. Rather, lower MCP production and efficiency is related to chemical and physical aspects of the diets. For chemical aspects, high concentration of secondary metabolic compounds, particularly tannins, in chromolaena (Aro *et al.*2009; YM Mulik, unpublished data) can bind to protein in the diet and makes it indigestible in the rumen (Van Soest 1996) hence reducing quantity to nutrients available for microbial growth. The physical

aspect that contributes the low  $E_{MCP}$  in present experiment was the particle size. Grinding of the feed stuff increased outflow rate of feed from the rumen as reported by Poppi *et al.* (1980) hence resulted in low rumen microbial growth.

## Conclusion

It can be concluded that *C.odorata* can potentially be utilized as a cheap protein source for fattened cattle when provided up to 40% in the total diet, but might have a negative effect when the levels raised above the current level since it shows a diminishing trend in important variables such as intake, digestibility and rumen function.

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# Substitution of Fish Meal by Cricket or *Indigofera* sp. Shoot Leaf Meal to Evaluate Protein Balance of Japanese quail (*Coturnix japonica*)

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## Abstract

Crude Protein content of Cricket meal and *Indigofera* sp. shoot leaf meal is relatively high, which are 55.59% and 27-29%, respectively. In addition, both of those feeds have complete amino acids. Those feeds are potential and well utilized as animal protein and plant protein to substitute fish meal in Japanese laying quail (*Coturnix japonica*) ration. This research was conducted to determine the potency of Cricket meal and *Indigofera* sp. shoot leaf meal to substitute fish meal in quail ration and to evaluate the protein balance. Thirty Japanese quails, at 35 weeks old were used in this study and kept in individual cages (two quails in each cage). A Completely Randomized Design was applied with three treatments and five replications. The treatments were R0 (diet contained fish meal), R1= substitution of fish meal with cricket meal, R2= substitution of fish meal with *Indigofera* sp. shoot leaf meal), based on dry matter requirement. Variables measured were feed intake, crude protein (CP) intake, meat and egg protein retention, metabolizable protein (MP), and protein utilization efficiency. The result showed that CP consumption and MP in R2 were higher ( $P < 0.05$ ) than other treatments, meanwhile feed intake, protein retention, protein utilization efficiency were same in all treatments. In conclusion cricket meal as animal protein source and *Indigofera* sp. shoot leaf meal as plant protein source could substitute fish meal utilization in laying quail ration without any negative effect to the protein balance.

Keywords: cricket meal, *Indigofera* sp. shoot leaf meal, Japanese laying quail (*Coturnix japonica*), protein balance

## Introduction

One thing which has a great importance for the commercial production of all avian species is the determination of nutritional requirements because diet represents the most burden some part of production and is perhaps the main factor that determines whether birds will grow to their maximum genetic potential. The nutritional requirements of quail are still poorly defined, conflicting, and are often based on extrapolations from requirements of other species of birds, or are obtained from foreign literature such as the NRC (1994). Giving high quality feed is known about the efficiency of energy utilization in regards to the deposition of protein and fat for growth in quails. Feed cost became one of constrain in improving quail productivity. Fish meal was a feedstuff containing high crude protein at a high price per protein percentage. Cricket meal and *Indigofera* sp. were feedstuffs containing high level of crude protein with 55.59% and 27-29% of crude protein, respectively. In addition, both of those feeds have complete amino acids. Those feeds are potential and well utilized as animal protein and plant protein to substitute fish meal in Japanese laying quail (*Coturnix japonica*) ration. Therefore, this study aimed to investigate the potency of Cricket meal and *Indigofera* sp. shoot leaf meal to substitute fish meal in quail ration and to evaluate the protein balance.

## Materials and Methods

### Laying Quail Raising

Thirty laying quail (*Coturnix coturnix japonica*) at the age of fifteenth weeks which have average weight 140 g were used in this research. They were divided in three kinds of treatments and five replicates. Quails were kept in the individual cages where two birds located in each age as a replicate. The cage used in this study has dimensions of 60 cm x 40 cm x 20 cm completed with feeder and manual drinker in each cage. The following treatments were tested to laying quail in each cage:

R0 : basal diet contained fish meal

R1 : basal diet substituted by cricket meal

R2 : basal diet substituted by *Indigofera* sp. shoot leaf meal

All treatments above were formulated based on Lesson and Summer (2008) to meet the nutrient requirements of 15 weeks old of laying quail (Table 1). Allocation of treatments into experimental units was based on completely randomized design.

Table 1. Composition of laying quail diet

Ingredients	R0	R1	R2
Yellow corn	51.95	51.95	49.04
Rice bran	4.5	2.95	0
Soybean meal	23	23	22.5
Fish meal	8	0	0
Cricket meal	0	8	0
<i>Indigofera</i> sp shoot leaf meal	0	0	15
Palm Oil	4.5	4.5	4
DCP	0.99	1.5	1.5
CaCO <sub>3</sub>	6.45	7.2	7.2
NaCl	0.25	0.4	0.4
Premix	0.2	0.2	0.2
DL-Methionine	0.16	0.3	0.16
Total	100	100	100
Nutrien contents			
Energy (kcal/kg)	2966.35	2976.81	2954.87
Crude protein (%)	18.30	18.76	18.00
Crude fat (%)	6.97	7.91	7.47
Crude fibre (%)	2.61	3.21	4.90
Methionine (%)	0.54	0.52	1.18
Lysin (%)	1.29	0.86	0.82
Sistein (%)	0.33	0.24	0.23
Methionine + Sistein (%)	0.87	0.76	0.62
Linoleic acid (%)	1.30	1.13	1.02
Ca (%)	3.24	3.10	3.12
P (%)	0.68	0.45	0.45
Na (%)	0.18	0.20	0.19
Cl (%)	0.24	0.28	0.28

## Procedures

Laying quail were fed by three kind of diets for four weeks with two times daily feeding (i.e 6 am and 4 pm). Each bird was fed 50 g per day and drink 300 mL. Total feed consumption was measured in the end of week. On the other hand, total water consumption was measured everyday. In the end of period, quail manure were collected to measure the true nutrient requirement of laying quail at 15 weeks old. The following are is the other variables measurement :

1. Protein consumption
2. It measured by feed consumption multiplied by protein content of the diet
3. Protein retention in egg
4. Proten retention in quail meat
5. Protein utility, which measured by dividing protein consumption and total retention
6. Protein excreta

## Data Analysis

Data were analyzed by using one way analysis of variance (ANOVA), the difference of tretaments was analyze by using Duncan's multiple range test with ANOVA test revealed significant different at  $p < 0.05$  (Steel and Torrie 1991).

## Results and Discussion

Treatments were fed for four weeks to laying quail at 15 weeks old were increased protein consumption of basal diet substituted by cricket meal. The increase of protein consumption is possible affected by increasing of protein content of the diet which substituted by cricket meal, yaitu 18.76%, at the same time the protein content of basal diet was 18.3% and basal diet was substituted by *Indigofera* sp. shoot leaf meal was 18%.

Table 2. Feed consumption and protein consumption of laying quail

Treatment	Feed consumption	Protein consumption	Total protein retention	Efficiency of protein utility	Digestible protein
R0	18.159 ± 1.221	3.323 ± 0.223ab	1.151 ± 0.097	0.346 ± 0.008	51.581 ± 15.069
R1	18.786 ± 1.062	3.524 ± 0.199a	1.241 ± 0.115	0.354 ± 0.052	52.058 ± 12.983
R2	17.463 ± 0.414	3.143 ± 0.075b	1.177 ± 0.126	0.375 ± 0.042	39.282 ± 15.658

Means in the same column with different superscript differ at  $p < 0.05$

Result showed that consumption of protein of crickets meal treatment (R1) was 3,524 g/head/day. These results were consistent with studies which has been conducted by Widjastuti and Kartasudjana (2006), that the consumption of protein for quail 3.49 g /head/day. The higher the protein content of the ration and feed consumption enabled the high protein consumption. Protein content of the diet treatment of indigofera meal substitution (R2) was the lowest which was compared with the others, this appropriated with the results obtained in which the consumption of protein of indigofera meal treatment (R2) was the lowest than others. The higher consumption of protein, will be the more protein substance that are used by quail for his maintenance or for other body functions, such as production (for example the protein content of meat and eggs) and reproductive (Praseno, 2005).

The increase of protein consumption could increase the amount of nitrogen which was retained in its body. Nitrogen retention was the amount of nitrogen that was not secreted in excreta, calculated from the amount of nitrogen of feed which was consumed minus the amount of nitrogen in the excreta regardless of endogenous nitrogen which was derived from uric acid nitrogen, bacteria and intestinal mucosa exuviation (Djunaidi and Natsir, 2003).

The result of protein retention of all treatments could be seen in Table 2. Increasing of the amount of protein consumption were influenced significantly by all treatments. The total protein (nitrogen) of all treatments were same. Based on Wahju (1997), not all proteins which came in to the body could be retained but it depend on genetic factors and age of quail. In addition, crude fiber content of feed could affect nitrogen retention in the carcass of quail. It could be seen that the lowest protein retention was on R0 treatment (fish meal supplementation).

Low level of protein retention were not appropriated with the amount of protein that enabled to be digested by the quail (all treatments). Based on the Table 2, all treatments produced low protein digestibility of quail feed. During the research, there were a decline in performance, especially average daily gain (ADG). ADG decrease were caused by lower feed digestibility. Protein retention of R0 and R2 were lower than R1. R1 was a diet which contained a crickets meal, and crickets meal protein content was higher than Indigofera meal (R2). While R2 treatment could be caused by fiber of Indigofera meal (17.83%) (Simanuhuruk and Sirait, 2009). The existence of a high fiber diet could decrease diet digestibility and tolerance limit of fiber of poultry (4%). Besides high fiber content, Indigofera also contain antinutrition (tannin). Result which has been conducted that tannin of *Indigofera zollingeriana* shoot meal was 0.29%, while the tolerance limit of tannin in chicken feed at 0.26% (Kumar *et al.* 2005).

## Conclusion

It can be concluded that cricket meal as animal protein source and *Indigofera* sp. shoot leaf meal as plant protein source could substitute fish meal utilization in laying quail ration without any negative effect to the protein balance.

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# The Study of Jack bean (*Canavalia ensiformis*) Addition on the Performance of Rats as Animal Model

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## Abstract

The purpose of this study was to examine the addition of Jack Bean (*Canavalia ensiformis*) in the diet to performances of rats (*Rattus norvegicus*) as an animal models to predict post-ruminal absorption. This study used a randomized complete block design with 3 replication, each replication consisted of 7 rats/block from 105 male rats on the growth-phase with an average weight  $117.84 \pm 23.52$ , with 5 treatments of ration: R0 = control diet, R1 = R0 + 10 % Jack bean flour, R2 = R0 + 20 % Jack bean flour, R3 = R0 + 30 % Jack bean flour, R4 = control diet + GHR1000. Data were analyzed by analysis of variance (ANOVA). The tested parameter were feed consumption, daily gain, feed conversion ratio, and chemical quality of the meat as dry matter, protein, fat as well as protein and fat meat ratio. The results showed that addition of Jack bean flour as much as 20% in the diet did not affect the consumption diet, but increased daily gain 1.98 g/day ( $P < 0.01$ ) and produced the best feed conversion ( $P < 0.05$ ) and resulted 10 fold of protein and fat meat ratio ( $P < 0.01$ ). As conclusion, addition of 20% Jack bean in the diet of rats which as an animal models to predict the post-ruminal absorption gave the best performance of rats as well as its meat quality. This means, if Jack bean will be contained in cattle diet, as much 20% Jack bean should be absorbed in post-ruminal digestive of cattle to produce high performance and high quality of meat.

Keywords: jack bean (*Canavalia ensiformis*), meat quality, performance, post-ruminal digestive, *Rattus norvegicus*

## Introduction

Indonesia is a famous country that has abundant biological resources such as legumes. Jack bean (*Canavalia ensiformis*) is one of legumes that can be used as a source of protein. Protein content of jack bean is 28.51%, 2.57% fat and 5.75% fiber. Unfortunately, jack bean has not been used as source of protein and energy. Nowadays jack bean is only used as food alternative to soybean and untapped as source of energy and protein feed for livestock. Nutritional content in jack bean is possible to be used as feed source of protein and energy, but high nutrient content also causes problems, especially for livestock performance. Therefore, it is necessary to develop an innovation to treat jack bean for feed protein and energy source purposes.

This research aimed to evaluate the protein used for beef cattle both small and large ruminants derived from jack bean in increasing muscle mass. Therefore, this study was conducted in the rat as animal model to describe the post-ruminal absorption. The purpose of this study was to examine the addition of jack bean (*Canavalia ensiformis*) in the diet on performances of rats (*Rattus norvegicus*) as an animal models to predict post-ruminal absorption.

## Materials and Methods

The experiment used 105 male rats on the growth-phase with an average weight of  $117.84 \pm 23.52$  g. Feed was given in pellets every morning and evening as well as water provided ad libitum. This study used a randomized complete block design with 5 treatments and 3 replications, each replication consisted of 7 rats. The treatments of ration were: R0 = control (negative) diet, R1 = R0 + 10 % Jack bean flour, R2 = R0 + 20 % Jack bean flour, R3 = R0 + 30 % Jack bean flour, R4 = control (positive) diet, GHR1000, is a synthetic growth promoter). Data were analyzed by analysis of variance (ANOVA) and Duncan analysis. The parameters were feed consumption, average daily gain (ADG), feed conversion ratio (FCR), and chemical quality of the meat such as dry matter, protein, fat, protein and fat meat ratio. The composition of feed was presented in Table 1 and the nutrient content of diet was presented in Table 2.

Feed intake recorded daily by weighing the amount of given feed and the feed residual. Feed intake was calculated as :

$$\text{Feed intake (g)} = \text{The amount given feed (g)} - \text{Residual feed (g)}$$

Measurement of body weight of rats was carried out every week during maintenance. The average of daily gain (ADG) was calculated based on formula as follows:

$$\text{ADG (g/h/day)} = \frac{\text{Final body weight/week (g)} - \text{initial body weight/week (g)}}{7 \text{ days}}$$

The feed conversion ratio was calculated by the amount of feed intake divided by the average daily gain as follows:

$$\text{FCR (\%)} = \frac{\text{Daily Feed intake (g/day)}}{\text{Daily body weight gain (g/day)}} \times 100\%$$

Chemical analysis of meat refers to AOAC 1984. The calculation of the ratio of protein and fat meat is calculated by the amount of meat protein content divided by the total fat content of meat.

Table 1. Composition of feed

Feed Ingredients	The amount used (%)
Corn	56.32
Pollard	19.29
CGM	16.24
CPO	3
Tapioca flour	2.5
CaCO <sub>3</sub>	1.5
DCP	1
Premix	0.1
Salt	0.05

Table 2. Nutrient content of diet control

Material	Nutrient content (%)					
	DM <sup>1</sup>	Ash <sup>1</sup>	Crude protein <sup>1</sup>	Crude fat <sup>1</sup>	Crude fiber <sup>1</sup>	NFE <sup>1</sup>
Control diet	88.54	4.25	18.40	5.42	2.35	58.12

<sup>1</sup>Analysis by Laboratorium Sumberdaya Hayati dan Bioteknologi PAU IPB (2014)

## Results and Discussion

Data of rat performance such as feed consumption, average daily gain (ADG) and feed conversion ratio (FCR) were presented in Table 3.

Table 3. The performance of rats

Treatment	Variable		
	Feed intake (g/h /day)	ADG (g/h /day)	FCR
R0	13.97±0.03	1.09±0.12C	12.81±1.54b
R1	12.72±1.37	1.96±0.28A	6.48±0.97a
R2	13.15±1.14	1.98±0.35A	6.64±1.33a
R3	12.83±1.03	1.51±0.19B	8.49±1.37a
R4	13.33±0.83	1.53±0.21B	8.71±1.62a

Notes: R0 = control diet, R1 = R0 + 10 % Jack bean flour, R2 = R0 + 20 % Jack bean flour, R3 = R0 + 30 % Jack bean flour, R4 = control diet + GHR1000. ADG = averagedaily gain.FCR = feed conversion ratio. Means in the same column with different superscripts (small font) differ significantly (P<0.05). Means in the same column with different superscripts (large font) differ significantly (P<0.01).

To obtain the optimal production, the diet consumption should meet the nutritional requirement. In this study, addition of jack bean flour in rat diets (R1, R2 and R3) did not affect the feed consumption compared to negative control (R0) and positive control (R4). This phenomenon shown that addition of jack bean flour with different levels did not change the animal's palatability. Jack bean as well as soybean contains high protein that preferred by animal. Beside of fulfilling the protein requirement of animal, the bean also favourable to trigger the positive palatability. This is because the jack bean has a high nutrient content, such as protein, fat, fiber, and other nutritional content, and also carbohydrate content of 50.8% (Doss *et al.* 2011).

There was significantly different on daily gain of rats as well as FCR by addition of jack bean flour ( $P < 0.01$ ). As much 10 % and 20% of jack bean flour in diet had produced similarity in the daily gain of rats as well as in feed conversion rate. The highest concentration of jack bean in diet (30%) showed decreasing the daily gain or increasing FCR. The quality and quantity of feed are the two factors that affect feed conversion ratio (Schmittows 1992).

Results of the analysis of the chemical quality of the meat during the study, such as protein, fat and protein and fat meat ratio provide a highly significant difference ( $P < 0.05$ ). The chemical analysis of meat can be seen in Table 4.

Table 4. Chemical quality of meat

Treatment	Variable			P/F ratio
	DM (%)	Protein (%)	Fat (%)	
R0	23.32±0.37	74.74±0.76A	16.78±0.98C	4.46±0.21B
R1	27.12±0.66	65.23±1.48B	12.19±1.27B	5.39±0.68B
R2	21.72±1.45	74.96±4.06A	7.46±1.41A	10.18±1.38A
R3	22.26±0.23	79.22±1.20A	14.47±0.46B	5.48±0.26B
R4	23.65±1.56	78.99±3.94A	13.35±0.60B	5.84±0.55B

Notes: R0 = control diet, R1 = R0 + 10 % Jack bean flour, R2 = R0 + 20 % Jack bean flour, R3 = R0 + 30 % Jack bean flour, R4 = control diet + GHR1000; DM = dry matter; P/F ratio = protein/fat ratio. Means in the same column with different superscripts (large font) differ significantly ( $P < 0.01$ ).

The addition of Jack bean in diet significantly influenced the meat protein content ( $P < 0.05$ ). Protein content of meat obtained in this study was in ranges between 65.23%-79.22%. According to Anggorodi (1994), the largest component of the dry matter is a protein (75-80%). In this study, the meat protein might be deposited as a mass of meat proteins. Protein deposited into the meat is excess protein from protein absorbed and utilized by the body, for example for tissue repair (Jamilah *et al.* 2013). The fat content of meat in this study was ranged from 7.46%-16.78%. The lowest fat content was achieved in addition 20% jack bean ( $P < 0.05$ ). This is the positive result, because consumers tend to choose the meat with a low fat content. The highest of protein and fat meat ratio was achieved by addition 20% jack bean flour. Its ratio was 2.3 times larger than control. The feed source of protein can be used to produce high protein mass. This proves that the jack bean flour can be consumed by livestock, especially beef cattle as a source of protein ration to increase meat production with better quality.

## Conclusion

The addition of 20% Jack bean in the diet of rats as animal models to predict the post-ruminal absorption gave the best performance of rats as well as its meat quality. This means, if Jack bean will be included in cattle diet, as much 20% Jack bean should be absorbed in post-ruminal digestive of cattle to produce high performance and high quality of meat.

## Acknowledgment

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# The Effect of Herbs Supplementation on Egg Quality and Lipid Blood of Laying Quail (*Coturnix-Coturnix Japonica*)

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## Abstract

The study was aimed to evaluate the effect of herbs supplementation through drinking water on quality of egg and lipid blood of laying quail. This experiment uses 400 heads of quail females, aged 36 weeks. Quails were reared until 8 weeks. Quail ration containing 18% crude protein and 2950 kkal/kg metabolizable energy. Herbs supplementation was called "jamu ternak" use as much as 9 kinds herb (*Alpinia galanga*, *Curcuma domestica*, *Kaemferia galanga*, *Curcuma xanthorrhiza*, *Zingiber officinale* Rosc, *Allum sativum* Linn, *Cinnamomun zeylanicum*, molasses and EM4). All herbs mashed, filtered and fermented for 5 days. Feed and water were given ad libitum. The experiment using a completely randomized design 4 treatments and 2 replications and two samples each replication. Data were analyzed descriptively. The treatments consisted of herbs supplementation was given through drinking water; i.e with 0,30 ml, 60 ml and 90 ml per liter of water was given twice a week. The variables measured were quality of egg (percentage of yolk weight, albumin weight, egg shell weight and score color of yolk egg) and lipid blood of laying quail i.e cholesterol, LDL, HDL and Triglyceride. This result showed that the treatments produced yolk color score ranges from 6-7, 30-32% yolk weight, 48-53% egg white and 10-12% shell weight. Herbs supplementation increased blood cholesterol of laying quail if was compared with control (without herbal supplementation). Levels of triglycerides, HDL and LDL tend to increased or decreased in the level of 30ml, 60 ml and 90 ml per liter water of herbal supplementation.

**Keywords:** egg quality, herbs supplementation (jamu ternak), lipid blood, quail

## Introduction

Utilization of herbs for poultry is increasingly popular in the people. Various herbal preparations called "jamu" has been widely studied and applied in poultry farms. Herbs like (jamu) made according to the interests and functions that can be selected from one kind or several kinds of herb, among others: kencur, garlic, ginger, galangal, turmeric, ginger, and cinnamon. In addition to many properties, materials for the manufacture of herbal medicine is also easily available in the market and reduce the dependence of farmers on chemical drugs and can reduce the cost of purchasing the drug. In general, herbal plants (rhizomes, leaves, stems, roots, flowers and fruits) have active compounds such as alkaloids, flavonoids, tripenoid, glycosides and essential oils. The active compounds are useful components to maintain the freshness of the body and improving blood circulation (Zainuddin and Wibawan 2007). Zainuddin and Wibawan (2007) stated that farmers routinely use herbs such as turmeric, garlic and papaya through drinking water or mixed in the animal feed showed avoid the attack of bird flu. Herb Supplementation (jamu) consisting of nine types of herbs can increase stamina, productivity and disease resistance, while herbal medicine consisting of 5 types of herbs can prevent bird flu, antiviral and increase productivity. Poultry were given herb herbs supplementation (jamu) will increase endurance (health), productivity, feed efficiency, carcass quality of chicken, meat and egg aroma, as well as animal manure does not smell sting (Zainudin, 2013). the use of herbs in chicken growth phase results in increased body weight and lower feed conversion. Zainudin, D-year. The use of turmeric can prevent coccidiosis, CRD, immune. The use of garlic prevent aflatoxin, improve appetite, digestion and anti-bacterial. Galangal improves the appetite, stamina, and increased appetite. Effect of herbal supplementation (jamu) on blood lipids has not been known. Blood lipid can describe the absorption of fat in the body that can affect the cholesterol content of eggs so that blood lipid quail information needs to be known by administering herbal medicine in drinking water.

## Materials and Methods

### Animal and Diet

Four hundred laying quails 36 week sold were placed in 10 colony cages. Each cage filled with 40 quail. The quail was fed ration contained 18 % crude protein and 2950 kkal/kg metabolizable energy. The ration was formulated based on the composition of the ingredients to NRC (1994). Ingredient composition of laying ration are shown in Table 1. Nutrient content of laying quail ration are shown Table 2.

Table 1. Ingredient composition of laying quail ration

Ingredient	Total (%)
Yellow corn	53.00
Rice bran	4.00
Soybean meal	25.00
Fish meal	7.50
Crude Palm Oil	3.00
CaCO <sub>3</sub>	6.00
Dicalcium Phosphat	0.60
Salt	0.30
Premix	0.50
DL-Methionin	0.10
Total	100

Table 2. Nutrient content of laying quailration

Nutrient	Content
Dry matter (%)	88.57
Crude protein (%)	18.9
Crude fiber (%)	3.12
Ether extract (%)	5.24
NFE (%)	50.68
Ash (%)	10.63
Ca (%)	3.2
P (%)	0.68
NaCl (%)	0.18
Metabolizable energy (kkal/kg)	2950

Laboratory Analysis Results Nutrition and Feed Technology, Faculty of Animal Husbandry, Bogor Agricultural University (2012).

### Herbal Supplementation (Jamu)

Making the herbal supplementation (jamu) consists of kencur (*Kaempferia galangan* L) (750 g), bawang putih (*Allium sativum* L) (750 g), jahe (*Zingiber officinale* Rosc) (375 g), lengkuas (*Lengkuas galangan* Stunz) (375 g), kunyit (*Curcuma domestica*) (375 g), temulawak (*Curcuma xanthorrhiza* Roxb) (375 g), and kayu manis (*Cinnamomun burmanii* B) (187.5 g) which still fresh plus molasses 300 ml and EM4 (Effective Microorganisms 4) 300ml (Saenab *et al.* 2002). The process of manufacture of medicinal herbs are chopped and mashed in a blender, then filtering and herbal extract taken. Afterwards, herbal extract are added with as much as 30 liters of water and then put in a plastic drum size of 30 liters and sealed for incubation process for 5 days. Jamu stirred 5 minutes everyday during the incubation process. The herbs supplementation had active compound represented in Table 3.

Table 3. Analysis result of herbs supplementation (jamu)

Active compound	Result analysis
Alkaloid	-
Flavonoid	+++
Phenol Hydroquinon	+
Steroid	-
Triterpenoid	++
Tanin	+

<sup>1</sup>Laboratorium biokimia, Fakultas Matematika dan Ilmu Pengetahuan Alam, IPB (2012).

## Quail Rasing and Collection of Data

The laying Quail reared 8 weeks at the farm people. Feed and water were given *ad libitum*. Giving herbal done twice a week on Mondays and Thursdays. Egg collection is done once a day in morning day. Five eggs samples taken everyweek then broken down and measured each variable. Eggs were coded according to treatments and weighed. The data were analyze during descriptive analysis.

## Treatments and Variables

The treatments in this experiment are:

R0 (1) : Control (Without herbal supplementattion).

R0 (2) : Control ( Herbal supplementation 90 ml/l water are given once a week )

R1 : Herbal supplementation 30 ml/l water are given twice a week

R2 : Herbal supplementation 60 ml/l water are given twice a week

R3 : Herbal supplementation 90 ml/l water are given twice a week

## Variables of this experiment are :

Quality of egg: egg weight, yolk weight, albumen weight, egg shell weight (g and %)

Lipid blood : Triglycerides, Cholesterol, HDL-Cho, LDL-Cho

Blood samples were taken from two quails each replication. Quail blood taken from the wing using the syringe and the analysis of lipid blood.

## Results and Discussion

Giving herbs supplementation through rinking water given twice a week on aquail during maintenance 8 weeks resulted in egg weight, yolk weight, albumin of egg weight, egg shell weight and egg yolk colors cores did not differ significantly (Table 3). Active substances contained in herbal medicine is not served to increase absorption of nutrients, seen no difference in the quality of the quail eggs given the same rations. The existence of active substances curcumin did not caused the color yellow yolk increased. The egg quality are more influenced by the content of nutrients that enter the body nature quail. The ration was given to the quail content 18% protein and 2950 kkal metabolizable energy that enough the needs a composition of normal egg.

Table 3. Egg quality of laying quail were given herbal supplementation in drinking water

	Control (1)	Control (2)	Herbal Supplementation (Its given twice a weeks)		
	Without Herbal Supplementation	Its one times a weeks	30 ml/l	60 ml/l	90 ml/l
Egg weight g	10.45	10.68	10.31	9.96	10.54
Yolk weight g	3.17	3.43	3.26	3.17	3.30
%	30.31	32.07	31.62	31.81	31.30
Albumen weight g	5.44	5.22	5.46	5.21	5.68
%	52.15	48.78	53.06	52.41	53.87
Egg Shell weight g	1.19	1.12	1.21	1.03	1.08
%	11.39	10.46	11.82	10.34	10.28
Score of yolk color	6.87	6.89	6.49	5.94	6.50
Cholesterol of yolk egg % w/w	2.92	2.68	2.73	2.78	2.77

The effect of herbal medicine in the drinking water on blood lipid content shown in the Table 4. The effect of herbs supplementation in crease from 30 ml until 90 ml by admin is tering 2 times a week tend to lower cholesterol blood levels and HDL blood levels but the triglycerides blood and LDL blood were not tendency to increse or decrease. Speed cholesterol synthesis in the body and environment affect blood cholesterol. (Murray *et al.*, 2000), especially feed and feed additive such as betaine (Shenatmoko, et al. 2013) or starbio (Saleh, et. al., 2013) and turmeric or ginger (Swastike, 2012). The effect of herbal or feed additive have varying results depending on the dose and the number of active substances into the body.

According Swastike. 2012. The addition a mixture of turmeric and ginger from 1-3% can lower blood cholesterol, but the addition of flour or extract katuk leaf did not reduced blood cholesterol. Park *et al.* 2002. Represented results this experiment that the supplementation of rutin and tannic acid promoted the excretion of fecal sterols, there by leading to a decreased absorption of dietary cholesterol as well as lower plasma and hepatic cholesterol in rats.

Information variables of blood lipids can be used to predict cholesterol content meat and egg of quail. Wiradimadja and Rahmat (2011) represented that cholesterol levels of blood can be used to predict the levels of egg yolk cholesterol ( $r=0.89$ ) or meat cholesterol ( $r=0.87$ ). Cholesterol levels of meat and eggs increased with increasing blood cholesterol, but will reach a maximum on blood cholesterol levels above 700mg/dl.

Level of LDL and HDL blood result of the experiment tend decreased with the level of herb supplementation from 30 ml/l - 90 ml/l. Shenatmoko *et al.* 2013 represented that Betaine supplementation 0-0.21% in ration of 14 weeks old quail significant effect on levels of blood LDL (92.80-188.93 mg/dl) but not significantly different at the level of HDL (8.28-18.75 mg/ dl).

Table 4. Lipid blood of laying quail was given herb supplementation (jamu) through drinker water

	Control (1)	Control (2)	Herbal Supplementation (Its given twice a weeks)		
	Without Herbal Supplementation	Its one times a weeks	30 ml/l	60 ml/l	90 ml/l
Triglyceride % w/w	194,06 ± 9.2	257,07 ± 70.8	171.81 ± 78.5	263,76 ± 90.0	233,49 ± 65.7
Cholesterol % w/w	160.06 ± 67.2	199.38 ± 24.7	227.86 ± 30.2	194.89 ± 46.1	193.81 ± 29.6
HDL-Chol % w/w	105.83 ± 10.6	84.20 ± 8.7	111,42 ± 21.1	103,3 ± 42.8	91,30 ± 16.9
LDL-Chol % w/w	50.93 ± 19.0	63.77 ± 30.8	82.08 ± 27.9	38.84 ± 31.5	55,81 ± 23.1

Laboratory Analysis Results Nutrition and Feed Technology, Faculty of Animal Husbandry, Bogor Agricultural University (2012).

## Conclusion

Giving herbs supplementation (jamu) a dose of 30 ml/l - 90 ml/l with twice administration in a week, does not affect the quality of quail eggs. Giving herbs with a dose of 30 ml/l - 90 ml/l to twice a week administration tends decreased cholesterol and HDL of quail laying blood.

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# Feed Intake, Weekly Gain and Feed Conversion of Growing Goats Fed Protected Fatty Acid

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## Abstract

An experiment was carried out to study response of growing goats fed protected fatty acids (PFA). Sixteen post weaning goats ( $\pm 4$  mo) and weight ( $\pm 14$  kg) were allocated to 4 treatments including  $T_1 =$  *Gliricidia sepium*,  $T_2 = T_1 + 200$  g PFA,  $T_3 = T_1 + 250$  g PFA, and  $T_4 = T_1 + 300$  g PFA, arranged according a completely randomized design. The results showed that there were significant differences ( $P < 0.05$ ) between treatments in feed intake, average weekly gain, and feed conversion. The feed intake and weekly gain of  $T_4$  were significantly ( $P < 0.05$ ) higher than those of  $T_1$ ,  $T_2$  and  $T_3$ , but feed conversion of  $T_4$  was the lowest. It is concluded that feeding protected fatty acid 300 g/head/day is the best treatment to improve growing goat performance.

Keywords : feed intake, weekly gain, feed conversion, growing goats, protected fatty acid

## Introduction

Growing goats during its active growth phase with traditional feeding practices (by grazing) could only achieve 0.40 - 0.50 kg average weekly gain, and need 9 - 12 months to attain body weight at 20 - 20 kg. Goats require high energy feed to support faster growth.

Protected fatty acid can be used as energy source for goat because it can increase energy content of diet and improve growth. Fats in ruminant diet can improve energy efficiency as the long chain fatty acid can be used directly in metabolic pathway of fat synthesis. Bhatt *et al.* (2011) showed that fat as energy source in proper form is prerequisite for improving pre- and post- weaning growth. Calcium salt of fatty acid (Ca-FA) prepared from rice bran oil had stimulated feed intake leading to higher total dry matter intake, improved body weight gain and feed conversion in lambs (Bhatt *et al.*, 2013). Therefore, the present experiment was planned to study the effect of supplementation with rice-bran protected fish oil as protected fatty acid on feed intake, average weekly gain and feed conversion.

## Materials and methods

The experiment was carried out at Animal Science Laboratory, Animal Science Faculty, UHO, Kendari. The climate is typical hot and semi-arid. The study was initiated in September 2013 and ended in December 2013. During the experiment, ambient temperature ranged from 30°C to 32°C.

## Feeding Treatment

Sixteen post weaning goats ( $\pm 4$  month olds) with the body weight  $\pm 9$  kg, were allocated to 4 treatments :  $T_1 =$  roughage,  $T_2 = T_1 + 200$  g PFA,  $T_3 = T_1 + 250$  g PFA, and  $T_4 = T_1 + 300$  g PFA. The goats were offered roughage after giving PFA in separate feeders. Feed was offered twice a day. The experiment was conducted until the goats reached six month olds ( $\pm 2$  months). Fish oil as wastes of tuna fish processing was used as the oil source. Protected fish oil (PFA) was prepared by mixing the warmed oil (1500 ml) with extract solution of *Spondias dulcis* forst leaf (300 ml) which were then stirred (10 min). This mixture was then added and mixed with rice bran (1:5 fish oil and rice bran on weight basis); the rice-bran protected fish oil was subsequently dried in oven at °C for min. Chemical composition of roughage and PFA is shown in Table 1.

Table 1. Chemical composition of roughage and PFA

Chemical composition (%)	Roughage	PFA
Moisture	67.10	8.44
Ash	9.70	11.11
Crude Protein	7.50	14.98
Ether Extract	2.20	9.96
Crude Fiber	25.50	12.99

### Sample Collection

Roughage and PFA samples were collected once a week by weighing the offered and the residue. Each samples collected every week were mixed thoroughly and pooled after drying for dry matter analysis and chemical composition.

### Variables measured in growth trial

Variables measured were : feed intake, body weight and feed conversion. Feed intake and body weight were measured every week, feed conversion was calculated from feed intake and body weight data.

### Statistical Analysis

Data were subjected to one way analyses of variances using SPSS base 16.0 with mathematic model as follows :

$$Y_{ij} = \mu + T_i + E_{ij}$$

where  $Y_{ij}$ : observation mean,  $\mu$ : general mean,  $T_i$ : effect of dietary treatment,  $E_{ij}$  : random error.

## Results and Discussions

Feed intakes of growing goats given PFA increased with the levels of PFA (Table 2), with the highest was obtained by giving T4 ( $P<0.05$ ). Feeding goat with T1 produced the lowest average weight gain, supplementing T1 with PFA increased the weight gain with the highest was obtained with T3 and T4 ( $P<0.05$ ). Feeding T3 and T4 also produced the lowest feed conversion ratio which was then increase with T2 and T1 ( $P<0.05$ ).

Table 2. Feed intake, weekly gain and feed conversion of growing goats fed protected fatty acids for 8 weeks

Variables	T1 (0 g PFA)	T2 (200 g PFA)	T3 (250 g PFA)	T4 (300 g PFA)
Dry matter intake (kg)	4.58 <sup>d</sup>	5.86 <sup>c</sup>	6.19 <sup>b</sup>	6.48 <sup>a</sup>
Average weekly gain (kg)	0.40 <sup>c</sup>	0.90 <sup>b</sup>	1.44 <sup>a</sup>	1.42 <sup>a</sup>
Feed conversion ratio	10.63 <sup>a</sup>	7.61 <sup>b</sup>	4.33 <sup>c</sup>	4.56 <sup>c</sup>

a,b,c Means with different superscript within the same raw are different ( $P<0.05$ ).

Results of the present experiment indicated that feeding PFA had positive effects on feed intake, average weekly gain and feed conversion ratio. These results were not in agreements with the results of De Souza *et al.* (2014) and Titi (2011) in which feeding CaS-FA as PFA had no effect on body weight gain and intake of total mixed ration (IMR). The performance of male Iranian native Mahabadi goat were not affected by giving fish oil (Najafi *et al.*, 2012). A similar result was also obtained by Karami *et al.* (2013) that supplementation with palm oil or canolla oil had no effect on performance of feedlot goat. Marinova *et al.* (2007) also demonstrated that fish oil supplementation did not cause significant changes in feed intake and average daily gain of local sheep breed. Adding protected fish oil and protected palm oil in ration did not cause differences in dry matter intake of goats.

The positive effects of PFA in the present experiment were in accordance to the results of Bhatt *et al.* (2013) and Al-Dabbas *et al.* (2011). Ca-soap rice bran oil supplementation in diet at 2.5% increased dry matter intake of lamb, improved body weight gain and feed conversion ratio (Bhatt *et al.*, 2013). Adding 3% of dry fat in ration was capable of increasing body weight changes (Al-Dabbas *et al.*, 2011).

## Conclusion

Protected fatty acid (PFA) supplementation can improve total feed intake, total weekly gain, and feed conversion during 8 weeks treatment with the best PFA level was 250 - 300 g/head/day.

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# Nutrient Profile and *in vitro* Digestibility of Fresh and Ensiled Cassava in Swine

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## Abstract

Market availability and price of conventional feedstuffs are variable, making imperative to explore alternative feedstuffs. Cassava (*Manihot esculenta*) is a starchy tuber that can be a potential feedstuff for swine. Three sample type (combinations of cassava parts): 100% tubers (T100), 50% tubers and 50% leaves (T50), and 25% tubers and 75% leaves (T25) were ensiled over three periods [fresh (M0), ensiled for two (M2) and three (M3) months]. Samples were analyzed for nutrient profile. *in vitro* digestibility of samples were determined using a 3-step enzymatic assay. Gross energy (GE) content ranged from 3838 to 5013 kcal/kg DM. The pH content ranged from 3.73 (T100-M2) to 4.23 (T25-M2), and dropped in respective combinations in M3 samples. DM digestibility was higher ( $P<0.05$ ) in T100 (87.6%) than T50 (72.6%) and T25 (50.9%). DM digestibility of M0 (76.1%) was higher ( $P<0.05$ ) than M2 (67.7%) and M3 (67.3%). GE digestibility was higher ( $P<0.05$ ) in T100 (86.9%) than T50 (70.0%) and T25 (51.6%), while GE digestibility of M0 (74.9%) was higher ( $P<0.05$ ) than M2 (66.7%) and M3 (66.7%) silage. There was an interaction ( $P<0.05$ ) between sample type and period of ensiling for both DM and GE digestibility. In conclusion, increasing leaves in samples decreased nutritional value of cassava silage but digestibility was still at a reasonable level. Thus, ensiling the combination of tubers and leaves of cassava can be useful to supply enough feedstuffs for swine. However, ensiling period needs to be considered with combination of tuber and leaves.

Keywords: alternative feedstuff, cassava silage, energy, *in vitro* digestibility, swine

## Introduction

Market availability and price of conventional feedstuffs like corn and soybean meal used in swine feeding are variable. Thus, it is imperative to explore alternative feedstuffs which can either completely or partially replace these feedstuffs to ensure the sustainability of the swine production. Use of alternative feedstuffs in swine diets can be optimized by characterizing their nutrient profile and digestibility. Cassava (*Manihot esculenta*) is a starchy tuberous plant which can serve as a potential alternative feedstuff for swine (Ravindran, 1995). Cassava roots are a good source of energy while leaves are rich in protein, vitamins and minerals (Montagnac *et al.*, 2009). Thus, combination of both roots and leaves can be a potential alternative feedstuff for swine. The objective of this study was to determine the nutrient profile and *in vitro* digestibility of cassava (*M. esculenta* var Crantz) ensiled over 2 periods as compared to fresh cassava samples.

## Materials and Methods

Three cassava sample type [combinations of cassava parts, 100% tubers (T100), 50% tubers and 50% leaves (T50), and 25% tubers and 75% leaves (T25)] were ensiled over three periods [fresh (M0), ensiled for two (M2) and three (M3) months]. The fresh samples were ground and analyzed for nutrient profiles including dry matter (DM), crude protein (CP), crude fat, acid detergent fiber (ADF), neutral detergent fiber (NDF), and gross energy (GE) contents following the standard procedures (AOAC, 2006). Starch content was determined using test kit (Megazyme International, Ireland; AOAC 996.11). The pH in silage samples was measured using a digital pH meter (SymPHony, VWR International, USA). Silage samples were freeze dried and analyzed following the same procedures. A 3-step enzymatic technique (Boisen and Fernandez, 1997) was used to get the *in vitro* apparent total tract digestibility of nutrients (DM and GE) in swine. The means of the nutrients digestibility were compared statistically using Proc MIX model of SAS 9.1 software and declared significantly different at  $P<0.05$ .

## Results and Discussion

There was wide variation in nutrient profile and *in vitro* nutrient digestibility of samples, both due to type and ensiling period. The pH of the silages ranged from 3.73 (T100-M2) to 4.23 (T25-M2), and dropped in respective combinations in M3 samples due to increased acid production from fermentation over time. The lower pH in the intestine may have potential gut health benefit to pig (Jha and Berrocoso, 2015). The GE content ranged 3838 to 5013 Kcal/kg DM of sample. Crude protein content increased with increasing inclusion of leaves (T25>T50>T100) as protein content in leaves is higher than tuber of cassava. Also, there was increased CP content with ensiling as the carbohydrates are fermented making the other nutrients more concentrated on DM basis. No effect of ensiling was seen in the fat content. Starch content ranged from 14.7 to 65.2%, and decreased as ensiling progressed (M0>M2>M3) as part of starch were utilized by microbes for fermentation over period. With increasing the leaves proportion in the mix, there was increase in the ADF and NDF content, which in turn, had negative effect on both DM and GE digestibility. It was expected as the leaves are richer in fiber content than tuber which is less digestible by swine. The DM and GE digestibility was significantly ( $P<0.001$ ) affected by both sample type and ensiling period. Also, there was an interaction ( $P<0.001$ ) between sample type and ensiling period for both DM and GE digestibility.

Table 1. *In vitro* apparent total tract digestibility of combination of cassava parts (Type) ensiled over different duration (Period) in swine

Variables	Type			Period			SEM	P-value		
	T100	T50	T25	M0	M2	M3		Type	Period	Type × period
DM digestibility	87.6	72.5	50.8	76.1	67.7	67.2	0.484	<.0001	<.0001	<.0001
GE digestibility	86.9	69.9	51.6	74.9	66.7	66.7	0.342	<.0001	<.0001	<.0001

\*All data is expressed on a dry matter basis

## Conclusion

Increasing leaves in samples decreased nutritional value of cassava silage, but digestibility was still at reasonable level. Ensiling resulted in a reduction of the CP, NDF and ADF contents compared with the fresh cassava samples. Thus, ensiling the combination of tubers and leaves of cassava can be useful strategy to supply enough feed for swine. However, ensiling period needs to be considered with combination of tubers and leaves. The increase in ensiling period decreased pH of the silages, such silage may provide gut health benefit, in addition to providing energy and other nutrients to swine.

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# Effect of Combination Silkworm Pupae Meal and Garlic Meal on Blood Profiles, Visceral Organs and Carcass Yield of Broiler Chicken

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## Abstract

*Silkworm pupae (*Bombyx mori*, L.) meal is a potential alternative feedstuff as protein source (50.52%) that can replace fishmeal utilisation in broiler ration. Garlic (*Allium sativum*, L.) meal has antimicrobial compound that could improve broiler growth performance. This experiment was aimed at investigating the effect of utilisation of silkworm pupae meal and garlic meal and its combinations on blood profiles, visceral organ weight and carcass yield of broiler chicken. A total of 180 DOC were randomly allocated into six treatments with three replications for each with 10 chicks per animal unit. The experiment were: P0 (Ration with fishmeal); P1 (P0 ration + 2.5% garlic meal); P2 (Ration with silkworm pupae meal replacing 25% of fishmeal); P3 (P2 ration + 2.5% garlic meal); P4 (Ration with silkworm pupae meal replacing 50% of fishmeal); and P5 (P4 ration + 2.5% garlic meal). Replacing fishmeal with silkworm pupae meal (P2 and P4 rations) has lowered plasma triglyceride and total protein ( $P < 0.05$ ). Combinations between silkworm pupae meal and garlic meal affected plasma glucose levels ( $P < 0.05$ ). However, there were no significant effects of treatments on hematology variables, carcass yield and visceral organ weights, except for spleen and abdominal weights. In conclusion, the treatments did not produce any negative effects on blood profiles, carcass and visceral organ yields. Silkworm pupae meal can be used as an alternative protein source up to 50% in replacing fishmeal, either without or with garlic meal addition, in broiler ration. An advantage of using silkworm pupae meal is producing healthier broiler chicken with less abdominal fat and lean carcass. Keywords: blood profiles, carcass, garlic meal, silkworm pupae meal, visceral organ.*

## Introduction

Increases in the demand for livestock products are expected to continue causing increases in feed requirements. Limitation in natural resources, climate change and competition between food - feed - fuel will decrease the availability of some conventional feed ingredients such as fishmeal. Requirements for fishmeal for Indonesian feed industry are fulfilled by importing the products from abroad. Indonesia imported as much as 56,046 tons of fishmeal in 2013 with the value was US\$ 69,221,000 (Ministry of Maritime and Fisheries 2914). Quality and quantity of fishmeal will affect productivity of poultry industry as fishmeals are used as the main protein source. To reduce the quantity of fishmeal in poultry feed, it is necessary to search for new feeds as protein sources as alternative feeds. Insects have potentials as alternative feed protein source because insects have good nutrient contents and similar amino acid profiles to those of fishmeals.

Silkworm pupae (*Bombyx mori*) is a waste product of rearing silkworms in silk production (sericulture). This waste product is rich in protein and can be used as alternative feed protein source to fishmeal. The centre of sericulture in Indonesia are located in South Sulawesi, West Java and Central Java with its potential production at about 1 000 tons of cocoons per year (Directorate of Social Forestry 2006). Silkworm pupae meal contained 58.28% crude protein, 28.93% crude fat (Mangisah *et al.* 2002), and 2 900 kcal kg<sup>-1</sup> of metabolizable energy (ME) with methionine and lysine contents were, respectively, 2.9% and 6.8% (Ravindran and Blair 1993). Use of silkworm pupae meal to substitute 50% of fishmeal did not cause any negative effects on broiler performance (Konwar *et al.* 2008). Substitution of fishmeal up to 100% with silkworm pupae meal, either fermented or not, produced a better feed conversion ratio on broiler (Rao *et al.* 2011).

Feed additive such as herbs can be added into ration to improve livestock products that can be consumed safely by human. Garlic (*Allium sativum*, L.) is a herbal plant containing allyl sulfide compounds (diallyl disulfide, diallyl sulfide and diallyl trisulfide) that have antioxidant effect (Santosh *et al.* 2013). The addition of 2.5% garlic meal in broiler diet improved feed conversion and carcass percentage (Suharti 2004) and increased the liver and spleen weights increasing the immunity of broilers (Wiryanawan *et al.* 2005).

This experiment was aimed at investigating the effect of dietary silkworm pupae meal and garlic meal and its combinations on blood profiles, visceral organ weight, and carcass percentage of broiler chicken.

## Materials and Methods

The experiment used 180 *Ross* strain day old chicks which were divided into six treatments in a Completely Randomized Design 3 x 2 factorial with 3 replications and 10 chicks for each replication. Factor A was silkworm pupae meal level substituting fishmeal in ration (0%, 25%, and 50%), factor B was garlic meal level in ration (0% and 2.5%). The treatments were: P0 (Ration with fishmeal); P1 (P0 ration + 2.5% garlic meal); P2 (Ration with silkworm pupae meal replacing 25% of fishmeal); P3 (P2 ration + 2.5% garlic meal); P4 (Ration with silkworm pupae meal replacing 50% of fishmeal); and P5 (P4 ration + 2.5% garlic meal). Nutrient composition of experimental rations for broiler starter (1-21 days) and finisher (22-30 days) were determined based on Leeson and Summers (2005). Variables observed were number of erythrocyte and leucocyte, blood glucose, triglyceride, total protein, percentage of carcass yield, and percentage of visceral organ yield. Data were analysed with analysis of variance (ANOVA); the significant data were then tested with Duncan Multiple Range Test (Steel & Torrie 1993).

## Results and Discussion

Result showed that utilisation of silkworm pupae meal replacing fishmeal, garlic meal, and its combination did not affect numbers of erythrocyte and leucocyte (Table 1). Blood profile could be used to describe the animal physiology status, and physiological disorder may change the blood profiles (Guyton & Hall 2010). Blood cell synthesis within the body required amino acid as substrate. Silkworm pupae meal contained complete and balanced essential amino acid which were similar to those found in fishmeal (Sanchez-Muroz *et al.* 2013). Use of silkworm pupae meal to replace fishmeal did not cause any changes in blood cell synthesis, the synthesis still occurred normally. Glutamine and glycine were amino acids playing positive roles in maintaining ATP levels in erythrocytes of broilers. Silkworm pupae meal contained 11.1% glutamine and 4.2% glycine (Tomotake *et al.* 2010). These amino acids were able to increase erythrocyte function. The effect of garlic was in association with sallicin as antibacterial compounds that could protect against Gram positive and Gram negative bacteria. This could be expected to boost the immune system (Wang *et al.* 2011) and caused the body to produce less leucocyte to fight bacterial infections. Garlic was also known to contain gurwitchrays that stimulated body cell growth and had rejuvenating power to all body functions including blood cells and caused synthesis of erythrocytes more rapidly (Santoso 1988).

The results also showed that blood glucose was affected by interaction between silkworm pupae meal and garlic meal in ration (Table 1). Broiler blood glucose in control group was significantly higher ( $P < 0.05$ ) than other treatments. The reduction in blood glucose could be due to the bio-active components, *s*-allyl cysteine sulfoxide (SACS) that were found in garlic meal; this bio-active compounds stimulated the secretion of insulin to lower the blood glucose levels. The decrease in blood glucose levels could also be due to the interference of glucose absorption from food by SACS compound (Srinivasan 2005). These results were also supported by the result of Santhosha *et al.* (2013) indicating that garlic may increase plasma levels of insulin in lowering blood glucose levels such as that occurred in diabetic rats and rabbits. Reduction in blood glucose levels was also observed when silkworm pupae meal was used up to 50% to replace fishmeal in ration. These reductions were due to chitin that presented in silkworm pupa meal as much as 3.37% (Singhal *et al.* 2001). Chitin was a polysaccharide, constituent of the crustaceans exoskeletons and insects, that were included in crude fibre groups that could not be digested by monogastric animals. Chitin was a polymer of 2-acetamido-2-deoxy- $\beta$ -D-glucose bound through  $\alpha$ -D-1-4 glycosidic binding establishing a linear polymer with long chain without side chain (Lindsay *et al.* 1984). The presence of chitin will decrease digestion and absorption of saccharide groups.

Blood triglycerides level was lowered ( $P < 0.01$ ) and was affected by utilisation of 25% and 50% silkworm pupae meal replacing fishmeal. Higher blood triglyceride level in control groups (Table 1) showed that fat absorption from the gastro intestinal tract occurred as normally. The reduction in blood triglyceride as a result of silkworm utilisation indicated that fat absorption from the gastro intestinal tract was affected by chitin. Chitin was able to bind bile acids that prevented interaction between digestive enzymes with fat particles in the duodenum; this binding reduced fat digestion (Pilliang & Djojosoebagio 2006). Inhibition of fat digestion decreased triglyceride synthesis and absorption.

Table 1. Blood profiles and blood metabolites of 30 days broiler chicken

Variables	Garlic Meal (B)	Silkworm Pupae Meal (A)			Means
		0%	Replace 25% Fishmeal	Replace 50% Fishmeal	
Erythrocyte (10 <sup>6</sup> mm <sup>-3</sup> )	0%	2.71 ± 0.20	3.15 ± 0.98	3.04 ± 0.44	2.96 ± 0.58
	2.50%	2.74 ± 0.45	2.62 ± 0.21	2.64 ± 0.27	2.67 ± 0.29
	Means	2.73 ± 0.31	2.89 ± 0.70	2.84 ± 0.39	
Leucocyte (10 <sup>3</sup> mm <sup>-3</sup> )	0%	5.60 ± 1.82	5.40 ± 1.32	7.07 ± 0.42	6.02 ± 1.39
	2.50%	5.30 ± 0.61	5.03 ± 2.30	5.50 ± 0.56	5.28 ± 1.24
	Means	5.45 ± 1.23	5.22 ± 1.69	6.28 ± 0.96	
Glucose <sup>a</sup> (mg dL <sup>-1</sup> )	0%	276.41 ± 36.70a	234.30 ± 24.03ab	207.61 ± 11.09b	239.44 ± 37.61
	2.50%	213.27 ± 29.81b	248.59 ± 32.75ab	245.02 ± 16.07ab	235.63 ± 28.96
	Means	244.84 ± 45.72	241.45 ± 26.85	226.32 ± 23.92	
Triglyceride <sup>b</sup> (mg dL <sup>-1</sup> )	0%	159.50 ± 12.56	97.39 ± 18.39	118.12 ± 9.79	125.01 ± 29.96
	2.50%	143.56 ± 19.59	112.05 ± 23.02	91.97 ± 15.02	115.86 ± 28.14
	Means	151.53 ± 17.11A	104.73 ± 20.29B	105.05 ± 18.27B	
Total Protein <sup>a</sup> (mg dL <sup>-1</sup> )	0%	3.58 ± 0.55	3.43 ± 0.67	2.94 ± 0.30	3.32 ± 0.44
	2.50%	3.73 ± 0.73	3.44 ± 1.16	2.98 ± 0.26	3.39 ± 0.50
	Means	3.66 ± 0.40a	3.44 ± 0.46ab	2.96 ± 0.18b	

<sup>a</sup>) Numbers in the same row and column with different small letters were significantly different at 5% test level (Duncan multiple range test), <sup>b</sup>) Numbers in the same row and column with different capital letters were significantly different at 1% test level (Duncan multiple range test)

Blood total protein levels were significantly lowered ( $P < 0.05$ ) and were affected by utilisation of 50% silkworm pupae meal replacing fish meal (Table 1). Higher levels of silkworm pupae meal utilisation increased the amount of chitin presented in the ration, and this affected the utilisation of proteins from silkworm pupae meal (Longvah *et al.* 2011). This was because of the ability of chitin to bind the proteins, and the amount of chitin could determine the degree of hardness of insects cuticle. Chitin contained approximately 7% nitrogen (N-acetylated glucosamine polysaccharide). However, this nitrogen content was not available for poultry because the birds did not have chitinase, an enzyme that digested chitin; as a result, chitin was difficult to be digested by the birds (Ravindran & Blair 1993).

Table 2 shows that broiler carcass yields were not affected by utilisation of silkworm pupae meal replacing fishmeal, garlic meal and its combination. This means that utilisation of silkworm pupae meal up to 50% replacing fish meal did not produced negative effects on carcass yields and the level of 50% was the best level in replacing fish meal; this result was supported and in agreement to those found by Konwar *et al.* (2008) and Makkar *et al.* (2014). However, full replacement of fishmeal with silkworm pupa meal decreased chick performances. The present result indicates that silkworm pupae meal can be used as a protein feed source replacing the use of fishmeal, especially for the broilers raised in the areas of sericulture centre such as in South Sulawesi, West Java and Central Java. The use of silkworm pupae meal as one of poultry feedstuff in those areas has an advantage, that is : a good protein source which is easily available nearby in great amount with a cheaper price than fishmeal, and a reduction in feed cost in raising the broiler chicken in those areas.

The only variables affected by the use of silkworm pupae meal in replacing fishmeal were the spleen and abdominal fat percentages (Table 2). The effect of silkworm pupae meal utilisation in increasing of spleen weight was due to the amino acids of silkworm pupae meal were used for antibody synthesis. Control group broiler chicken had the highest abdominal fat percentage meaning that fat digestion and absorption occurred normally. The reduction in abdominal fat percentage with the use of silkworm pupae meal was in relation with the presence of chitin. This chitin affected fat digestion and absorption through its effects on blood triglyceride an total protein variables, as well as on blood glucose when it was combined with garlic meal (Table 1). These mean that using silkworm pupae meal in replacing fishmeal could produce healthier broiler chicken with less abdominal fat and lean carcass. All visceral organs weights were in the normal range. This means that the use of silkworm pupae meal did not influence the organ performances, and the nutrients of silkworm meal could be used to synthesis body cell and maintain the organs as good as fishmeal. These good effects of the use of silkworm pupae meal were also supported by the use of garlic meal that also contained antioxidant and antibacteria.

Table 2. Carcass and visceral organ yields of 30 days broiler chicken

Variables	Garlic Meal (B)	Silkworm Pupae Meal (A)			Means
		0%	Replace 25% Fishmeal	Replace 50% Fishmeal	
Carcass yield (%)	0%	61.31 ± 2.73	61.29 ± 1.97	61.04 ± 2.95	61.22 ± 2.24
	2.50%	59.61 ± 4.16	59.48 ± 1.29	59.51 ± 4.46	59.53 ± 3.12
	Means	60.46 ± 3.28	60.39 ± 1.79	60.28 ± 3.49	
Liver (%)	0%	3.65 ± 0.46	3.24 ± 0.22	3.34 ± 0.25	3.41 ± 0.34
	2.50%	3.65 ± 0.68	3.26 ± 0.06	3.30 ± 0.22	3.40 ± 0.40
	Means	3.65 ± 0.52	3.25 ± 0.15	3.32 ± 0.21	
Heart (%)	0%	0.63 ± 0.07	0.62 ± 0.07	0.62 ± 0.04	0.66 ± 0.05
	2.50%	0.71 ± 0.10	0.68 ± 0.05	0.62 ± 0.06	0.67 ± 0.07
	Means	0.67 ± 0.09	0.65 ± 0.06	0.62 ± 0.05	
Gizzard (%)	0%	2.35 ± 0.52	2.00 ± 0.07	2.43 ± 0.45	2.26 ± 0.40
	2.50%	2.41 ± 0.24	2.18 ± 0.23	2.52 ± 0.28	2.37 ± 0.27
	Means	2.38 ± 0.36	2.09 ± 2.18	2.48 ± 0.34	
Spleen (%)	0%	0.14 ± 0.01	0.21 ± 0.09	0.17 ± 0.02	0.17 ± 0.06
	2.50%	0.13 ± 0.04	0.19 ± 0.05	0.17 ± 0.01	0.16 ± 0.04
	Means	0.13 ± 0.03b	0.20 ± 0.07a	0.17 ± 0.01a	
Bursa fabricius (%)	0%	0.06 ± 0.01	0.06 ± 0.01	0.09 ± 0.02	0.07 ± 0.02
	2.50%	0.06 ± 0.02	0.08 ± 0.01	0.07 ± 0.02	0.07 ± 0.02
	Means	0.06 ± 0.01	0.07 ± 0.01	0.08 ± 0.02	
Abdominal fat (%)	0%	1.27 ± 0.38	1.04 ± 0.13	1.08 ± 0.10	1.13 ± 0.23
	2.50%	1.24 ± 0.13	0.79 ± 0.09	0.93 ± 0.14	0.99 ± 0.23
	Means	1.25 ± 0.25a	0.92 ± 0.16b	1.01 ± 0.14ab	

Numbers in the same row and column with small letters were significantly different at 5% test level (Duncan multiple rangetest)

## Conclusion

In conclusion, the treatments did not produce any negative effects on blood profiles, carcass and visceral organ yields. Silkworm pupae meal can be used as an alternative protein source up to 50% in replacing fishmeal, either without or with garlic meal addition, in broiler ration. An advantage of using silkworm pupae meal is producing healthier broiler chicken with less abdominal fat and lean carcass.

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**THEME E.**  
**ANIMAL GENETIC, BREEDING, AND REPRODUCTION**

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# Analysis of Captive Breeding Management of Silvery Gibbon (*Hylobates moloch* Audebert 1798)

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## Abstract

Captive breeding is aimed to intensively breed wild animals in the cage for biomedical research interests and to protect endangered species. The Primate Research Center of Bogor Agricultural University (PSSP-IPB) in collaboration with Taman Safari Indonesia (TSI) has established an ex-situ captive breeding of the Javan gibbon and has succeeded in breeding the species. Due to the limitation of cages at PSSP-IPB, the whole family of the Javan gibbon was moved to TSI, and therefore the research was conducted at TSI from February to July 2013. The objective of the research was to analyze the breeding management of the species. The results showed that the transfer of the Javan gibbon from PSSP-IPB to TSI had resulted in the changes of breeding management including cage system, environmental enrichment, feeding system, sanitation, and health control. Human resources and reproductive management in captivity of TSI and USDP-IPB did not significantly change since the animals had two nurses and one veterinarian, and the Javan gibbon in two other locations had already succeeded in delivering a baby.

**Keywords:** captive breeding management, ex-situ, *Hylobates moloch*

## Introduction

Captive breeding is an attempt to breed wild animals intensively in the cage. The breeding of primates in captivity has two objectives: to produce animals for biomedical research interests and to protect endangered wildlife species (De Mello 1991; Iskandar 2007). The Javan gibbon (*Hylobates moloch*) is one of the primates that belong to the category of endangered species in Indonesia since this animal can only be found in certain areas in West Java and Central Java (Roos *et al.* 2014).

One way to increase the Javan gibbon population in Indonesia is by carrying out captive breeding as has been done by Primate Research Center of Bogor Agricultural University (USDP-IPB) in Bogor. According to Thohari (1985) and Jarkasih (2005), breeding technology includes a number of aspects such as housing, food, reproduction, and health. Captive wildlife breeding is considered successful if the animal reproductive technology has been understood, and the animals in the captive breeding have bred successfully.

The Javan gibbon female in captivity at USDP-IPB from 2003 has successfully given birth to 5 young gibbons. The first offspring is OJ, the second is JLO, the third is OLA (Riendriasari *et al.* 2009), who died of bacterial infection, the fourth is OO, and the fifth is Ano (Nuraisah *et al.* 2011). As OJ was getting older and mature, he needed a spouse. If the animals were not separated, it was feared that in-breeding might occur between one child and another child, or even between a parent and a child. That is why, they were moved from USDP-IPB to Taman Safari Indonesia (TSI). This study aimed to obtain and analyze information on the management of the Javan gibbon captive breeding at USDP-IPB in the period 2003-2013 and post-transfer to TSI in 2013. There is still no information on the change in the Javan gibbon management after moving to a new breeding location. Therefore, it is necessary to conduct research on captive management of the Javan gibbon in TSI.

## Materials and Methods

The study of the Javan gibbon captive management was conducted from February to July 2013 in TSI, Bogor, West Java. The research was focused on the Javan gibbon family that were moved from USDP-IPB to TSI. They consisted of Ari (the father), Mimis (the mother), OJ (almost mature male), OO and Ano (both still very young). Meanwhile, JLO (almost mature female) was not observed since she was separately placed in an individual cage. The equipment used in this study included thermometer, digital camera of Casio brand, measuring tool, watch, and stationery.

The primary data on captive management were obtained not only from direct observation but also from the identification of breeding facilities as well as animal-caring methods in TSI, including housing, environmental enrichment, feed provision, health control, sanitation, human resources, and reproduction. The secondary data were obtained from the previous studies at USDP-IPB, among others, by Iskandar (2007) and Rahman (2011). In addition, the interviews with officials, documents, and relevant literature from a variety of sources had also contributed to the secondary data.

The data were analyzed descriptively in order to get treatment information showing a general overview of the state of captive management of the Javan gibbon. The aspects of the captive management analyzed included housing, environmental enrichment, feed provision, health control, sanitation, human resources, and reproduction.

## Results and Discussion

### Cage Management

There are two kinds of the Javan gibbon cages in TSI. The first cage is rectangular-shaped with an average volume of 78.804 m<sup>3</sup>/individual, and the second cage is similar to the natural habitat of the animal, which is located at the mountain slope where there are plenty of shade trees with an ambient temperature of around 16-25°C. Meanwhile, the Javan gibbon cage at USDP-IPB (Rahman, 2011) is divided into two rectangular-shaped enclosures with an average volume of 30.415 m<sup>3</sup>/individual, which are located in the middle of office and residential areas so that the condition is more noisy and hotter with an ambient temperature of about 20 -25°C. Both breeding locations have a partition made of stainless steel wire that separates the cages. The size of the cage in TSI is 2.5 times wider and the condition is more comfortable for the Javan gibbon to do their daily activities such as swinging, jumping, hanging, climbing up or down, etc.

### Environmental Enrichment

Environmentally, TSI management made an effort to enrich the cages of the Javan gibbon so that the conditions of the cages resemble their natural habitat. The types of enrichment in each cage of the Javan gibbon in TSI are the use of different materials with different numbers. The enrichment includes 2 wooden boxes, 2 wooden trunks, and 2 wooden seats; 1 plastic feed box, 2 aluminum water boxes, 1 rubber tire, and 1 piece of rope. In the meantime, at USDP-IPB (Rahman, 2011), the enrichment includes 2 wooden boxes and 2 wooden seats, 1 plastic feed box and 1 plastic water container, 1 aluminum water container, and 5 steel chains.

Table 1. Types and number of environmental enrichment elements in TSI and USD

Type of environmental enrichment	TSI	PSSP-IPB (Rahman 2011)
Number of types	7 types	6 types
Number of the same types	4 types (wooden box, seat, feed box, water box)	
Number of different types	3 types (rope, tree trunk, rubber, tire)	2 types (chain, water container)

Each type of the environmental enrichment has different functions, for example, a wooden box is used as a replacement for a tree to sleep for a rest, while the ropes, chains, and water containers are used to help the animals with swinging movements.

### Feeding

The feeding techniques in TSI are in the form of fruits, vegetables and feed additives (monkey chow) by means of separation or sorting the fresh feed from the rotten feed. Then the fresh fruit is cut into several parts (skins and seeds are not discarded), while the vegetables are cut on the hard parts of the stem and the roots. The food materials that have been cut are weighed so that the weight of the feed given is according to the number of individuals in each cage and the feed is put into a plastic basin.

Table 2 showed that TSI had more types of feed for the Javan gibbon (15 types) than USDP-IPB did (13 types). There were 10 feed types which were the same and there were different types of feed (5 types in TSI and 3 types in USDP-IPB). The Javan gibbon favorite types of feed in TSI and USDP-IPB were bananas, oranges, and *kangkung*, probably because of their sweet taste and soft texture, or maybe because the feeds have compatibility with the characteristics of the Javan gibbon that consume lots of fruits

that contain carbohydrates (banana), vitamin (orange), and fiber (*kangkung*). The types of feeds that the animals disliked both in TSI and in USDP-IPB were dominated by plant types because the feed types have sour taste, rough skin texture, and a lot of fiber, making it difficult for the body to digest.

Table 2. Feeding of the Javan gibbon in TSI and USDP-IPB

No	Type	TSI	USDP-IPB (Rahman (2011))
1.	Feeding time	Morning and afternoon	
2.	Number of feed types	15 types	13 types
3.	Feed of the same types	10 types (banana, apple, orange, salak, guava, monkey chow, long bean, spinach, <i>kangkung</i> , and <i>kemang</i> leaves)	
4.	Feed of different types	5 types (rambutan, mango, <i>kesemek</i> fruit, celery leaves, daun mede)	3 types (carrot, sawi, and green bean)
5.	Types of favorite feeds	banana, orange, and <i>kangkung</i>	
6.	Types of disliked feeds	celery, cashew leaves, long bean and mango	Green bean, carrot, and long bean.

### Health Control

TSI has more types of health support facilities for Javan gibbon than USDP-IPB does. There are 8 types which are the same and there are different kinds of support facilities (6 types in TSI and 2 types in USDP-IPB). The types of supporting facilities can be seen in Table 3 below.

Table 3. Types of the Javan gibbon health support facilities in TSI and USDP-IPB

No	Type of facility	TSI	USDP-IPB
1.	Number of facility types	14 types	10 types
2.	Types of the same facilities	8 types (individual cage, necropsy room, crematory room, operating room, pathology room, quarantine room, operational vehicle, and library)	
3.	Types of different facilities	6 types (animal hospital, room medicine, nursery, in-patient room, x-ray room, CR room (Computeres Rountsen))	2 types (microbiology room and immunology room)

To prevent bacteria or diseases from attacking the Javan gibbon in TSI, the officers wash the surface (skin) of feed with clean water, clean cages and provide adequate nutrition. In addition, the officers also clean their boots whenever they want to enter the cages by putting their boots in a container which already contains a mixture of water and disinfectant that is placed in front of the entrance (footbath) to prevent dirt (germs) from being carried over into the cages. The daily check of the Javan gibbon health in TSI and USDP-IPB is performed by a nurse twice a day i.e. in the morning and afternoon by checking the physical condition of the animals (sores, hair loss), appetite (weak), residual stool (feces and urine ) and cage facilities. If the Javan gibbon is sick, the nurse will contact the veterinarian to provide health care. In both captive locations, the Javan gibbon obtained the same health control.

### Sanitation

The cleaning of the Javan gibbon cages in TSI used a wet system (using water) once a day, in the morning by sweeping the remaining stool (feces, urine, and food scraps) and scrubbing it using water. The disinfectant is applied once a week prior to the feeding to prevent bacteria that can endanger the health of the Javan gibbon. These results differed from Rahman (2011) in USDP-IPB who stated that cage cleaning should be done with a wet system (using water) twice a day i.e. in the morning and afternoon.

### Human Resources (HR)

In the execution of daily tasks, the officers in TSI and USDP-IPB were accompanied by two nurses (senior high school graduates) and one veterinarian (S1 graduate) who has good skills in health care of the Javan gibbon with 10 working hours a day.

### Reproduction

In TSI, Ari and Mimis had 14 times mating, and on July 9, 2013 the mother gave birth to a baby. The time interval between mating activity and the birth of a baby in TSI was only 4 days before giving birth.

Iskandar (2007) stated that the birth interval between the first and the second child at USDP-IPB was about 14 months, whereas in TSI the birth interval between the fifth and the sixth child was 24 months. This shows that the older the mother, the longer birth interval. The birth interval between one child and another at USDP-IPB and in TSI was faster than the previous statement which mentioned 2-3 years (CBSG 1994; Campbell, 2008) and 3-4 years (Supriatna and Wahyono 2000). With the birth of the babies, the Javan gibbon breeding in both locations (TSI and USDP-IPB) was considered successful.

## Conclusion and Recommendation

The transfer of the Javan gibbon from USDP-IPB to TSI was followed by the changes in the management of cages, environmental enrichment, feeding, sanitation, and health control. These changes affected the behavior of the animals observed in moving, playing, sounding, browsing, eating, defecating, breastfeeding, and breastsucking. It is suggested to increase the Javan gibbon breeding techniques, especially during the care of the young and the mating method to accelerate population growth along with the management of adequate cage enrichment in terms of the area size and the existing facilities to avoid the appearance of abnormal behavior.

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# Phenotypic Variation in Male Local Chicken at Tapin Regency Using Significant Analysis

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## Abstract

*This aims of the research was to study the phenotype characteristic of male local chicken using significant, correlation and regression analysis in District of Tapin. Research was conducted in the District of Tapin, using 120 male local chickens. Observed variables, namely the tibia length, body length and body weight. Data were analyzed using MINITAB program. The results showed the analysis results of correlation with body weight, body length and tibia length. Regression was most influence on body weight that the age of 8 month of  $B = -707 + 108$  body length. Correlation of body weight and body size range male local chicken at the age of 5-8 month with significant ( $P < 0.05$ ) is very strong values contained in the body length of the age of 8 month 0.986; determination ( $R^2$ ) = 97.2%; regression  $B = -707 + 108$  tibia length. This correlation can be used as a reference for the selection of local chicken in the District of Tapin.*

*Keywords: correlation, male local chicken, phenotypic variation, significant analysis*

## Introduction

One source of protein of animal origin that is very easily available and widely known by the public, namely chicken. As a source of animal protein, chicken has advantages where meat and eggs are preferred community. The price is relatively more expensive than chicken. Several regions in Indonesia society chicken used in religious ceremonies, customs and pleasure so cultivation needs to be improved (Goddess and Wayan, 2012). Domestic poultry populations in South Kalimantan Province pata 2008 to 2013 consecutively tail 12,643,202; 12,911,052 tail; 13,702,575 tail; 12,847,604 tail, and the tail 9.98731 million (Anonim, 2013). While the domestic poultry population in North Tapin Tapin district districts in 2013 ie 120 320 tail (Anonim, 2013). The genetic information is required to determine the genetic quality of an animal that will be used as consideration in the selection or crosses. One of the basic research to explore the genetic information is phenotypic observations with measurements of morphology, as is done on chickens by Ojedapo *et al.*, (2012). This will help to efforts to increase the productivity of chickens. Productivity chicken is not enough just to feed improvement and maintenance management, but it is necessary to improve the genetic quality by maintaining the distinctive properties owned by the chicken. Each type of chicken has the typical characteristics of both size and shape. High productivity is an important factor in a farm because it determines the success or failure of these farms. Body measurements can be used to study the growth and development of the livestock. Cattle with greater bone size tend to grow faster and produce larger carcass pieces anyway, but the fact there is now a decline in the quality of village chickens in the community. In connection with the unfavorable conditions of the population, the improvement of the genetic quality of chicken needs to be done. Improvement of the genetic quality of livestock can be done in the selection and / or a cross. Quality improvement of animal genetic progress through selection is determined by the strength of inheritance of traits to be repaired (Warwick *et al.*, 1990). Further noted also that the weight has a relationship (correlation) with the real size of the body. Based on these reasons, this study was conducted to add information about the relationship of body weight and body measurements in domestic poultry. This study aims to determine the correlation of domestic poultry body weight and body size as the length of the tibia, shank length, body length, body height and chest circumference, as well as the size of the body where the relationship was highest in the age of 5 - 8 months.

## Materials and Methods

This research was conducted in the District of Tapin, South Kalimantan. This study carried out for 2 months until November to December 2014. Includes: preparation, implementation research, data processing

and reporting results. The research material is chicken male the age of 5-8 months from people cattle domestic poultry breeders in the area obtained from the District of Tapin 240 chickens chicken consisted of males. The tools used are scales, calipers, measuring record, spreadsheet questioner, stationery tools and a camera digital. The method used in this research is the survey, whereas the observed parameters, namely the appearance of free-range chicken male and female domestic poultry which consists of: weight, length of the tibia, shank length, body length, body height and chest circumference. Determine to the effect of the tibia length, shank length, body length, body height and body weight chest circumference with domestic poultry in the District of Tapin Correlation analysis, regression, determination and significant test with the program MINITAB 14.

## Results and Discussion

### Overview of Native Chicken in Tapin District

Many domestic poultry kept by farmers in the district of North Tapin Tapin district is the type of cruciferous bangkok domestic poultry, but the types of domestic poultry can not be ascertained due to free-range chicken at this point has experienced crossover of various types are not directional. The domestic poultry breeders belonging to maintain itself. In general, the origin of seeds obtained from a local free-range chicken with its own hatchery and buy seeds from other farmers. While building the average cattle sheds situated around the residence breeders with the maintenance system extensively. Feed given in the form of the free-range chicken from food waste (leftover rice, leftover vegetables, fish, etc.) and agricultural waste (bran, coconut pulp, pulp, etc.). Frequency of feeding an average of 2 times a day morning and afternoon.

### Overview of Results Measurement Body Weight (Tibia Length and Body Length)

Table 1. Mean and standard deviation body weight and body size native chicken males in District of Tapin

No	Sex	Age (month)	BW (gr)	TL (cm)	BL (cm)
1.	Male	5	1476.10 ± 202.19	11.21 ± 0.94	18.97 ± 1.97
2.	Male	6	1828.17 ± 289.50	12.28 ± 1.11	22.20 ± 1.83
3.	Male	7	1981.33 ± 391.73	13.08 ± 0.89	23.27 ± 2.65
4.	Male	8	2137.00 ± 356.17	13.37 ± 1.16	26.22 ± 3.24

Description: BW = Body Weight, TL = Tibia of Length, BL = Body of Length

The results of average and standard deviation of body weight and body size range chicken males from the age of 5-8 months obtain different values (Table 1). The average weight 5 months of age (1476.10 ± 202.19), 6 months (1828.17 ± 289.5), 7 month (1981.33 ± 391.73) and 8 months (2137.00 ± 356.17). One measure of the body of the tibia length 5 months of age (11.21 ± 0.94), 6 months (12.28 ± 1.11), 7 month (13.08 ± 0.89) and 8 months (13.37 ± 1.16). Value shows from the age of 5-8 months the higher value. In accordance with the opinion of Erwinda (2012) which states that, the size of the body is affected by age. Murtidjo (1989), the older cattle will change in size, shape/appearance and body composition. Body weight and body size domestic poultry average male shows the value that is not much different from the research Nugraha (2007) states the average weight range chicken that is 2405.141 ± 151.510 gr male. The length of the average body size Tibia 152.95 ± 10.24 mm in males. As well as research Mariandayani *et al.* (2013) stated the average body size shank length of 68.75 ± 5.14 mm male, chest circumference of 2.26 cm 17.70 + males.

### Correlation with Body Weight Body Size Native Chicken at Aage 5-8 Months

Table 2. Correlation native chicken male age 5-8 months

No	Sex	Age (month)	Tibia Length		Body Length		High Body	
			r	R (%)	r	R (%)	r	R (%)
1.	J	5	0.835	69.80	0.933	87.10	0.918	83.30
2.	J	6	0.944	89.10	0.911	83.00	0.966	93.40
3.	J	7	0.953	90.9 0	0.962	92.5 0	0.831	69.00
4.	J	8	0.952	90.60	0.986	97.20	0.811	65.70

Male domestic poultry have correlation values (r) is very strong and determination at the age of 5 months (ie r = 0.933 and R = 87.1%), 6 months (r = 0.973 and R = 94.7%), 7 months (ex. r = 0.962 and R = 92.5%), and 8 months (ex. r = 0.986 and R = 97.2%).

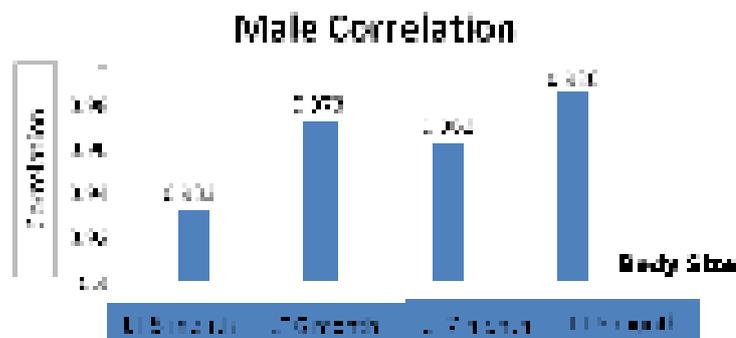


Figure 1. Correlation graphs native chicken male age 5-8 months

Correlation of body weight and body size range chicken males aged 5-8 months who has a very strong correlation values contained in the body length of the age of 8 months is 0.986. Then there is the age of 6 months height is 0.973, body length is 0.962 aged 7 months and 5 months of age body length is 0.933. Mansjoer (1981) examined the relationship of body weight to body size, which acquired a strong relationship between body weight to tibia length ( $r = 0.98$ ), with a long shank body weight ( $r = 0.98$ ) and body weight with a chest circumference ( $r = 0.95$ ). Erwinda (2012) which states that, the size of the body such as the length of the body is affected by age. Murtidjo (1989), the older cattle will change in size, shape / appearance and body composition.

## Conclusion and Suggestion

### Conclusion

Based on the results of research and discussion, it can be concluded that:

1. The male local chicken at the age of 5-8 months had average body weight and body size are different. Differences in body weight and body size domestic chicken because of differences in age, sex, type and amount of food consumed as well as genetic.
2. Correlation of body weight and body size range chicken male at the age of 5-8 months with a very strong correlation values contained in the body length of the age of 8 months, namely 0.986; Determination ( $R^2$ ) = 97.2%; BW regression equation =  $-707 + 108 \text{ BL}$  and significant ( $P < 0.05$ ). This correlation can be used as a reference selection on male domestic poultry in the district of Tapin in selection male local chicken.
3. Correlation of body weight and body size range chicken females at the age of 5-8 months with a very strong correlation values are at the age of 8 months chest circumference is 0.981; Determination ( $R^2$ ) = 96.1%; regression BW =  $-1\ 395 + 98.7 \text{ LB}$  and significant ( $P < 0.05$ ). This correlation can be used as a reference selection on female domestic poultry in the district of Tapin in selection male local chicken.

### Suggestion

The parameters used in this study only body weight, shank length, tibia length, body length, body height and chest circumference alone. To complement the data and determine the development of chicken, further research is necessary to add variables such as length of the back, chest width, beak length and the number of chickens that much. As well as in the study maintenance intensive observation at the same time.

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# Effects of Selection on the Efficiency and Variability of Sow Reproduction and Maternal Abilities

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## Abstract

The objective of this study was to estimate the effects of selection on the efficiency and the variability of reproductive and nursing performance of French Large White (LW) sows along their career (6 parities). LW sows were inseminated with stored frozen semen of LW boars born in 1977 and 1998, leading to 2 experimental groups of pigs (D7 and D8). A second generation was produced by inter se mating within D7 and D8 groups. Cross-fostering was practised shortly after birth to obtain mixed D7-D8 litters to investigate direct and maternal effects on piglet growth. Traits investigated included the number of piglets born in total (TNB) and born alive (NBA) per litter; litter weight at birth (LWB), sow weight loss from birth to weaning (SWL), weaning to first oestrus interval (WEI), individual and average piglet weight at birth and at 21 days of age. The data were analysed using mixed linear models. The variance of performance across parities was computed from mixed models residual values. All traits were significantly affected by selection, with an increase in litter size and weight (+0.12 and +0.09 piglet/year and +0.22 kg/year for TNB, NBA and LWB, respectively), a higher SWL (+0.52 kg/year) and a shorter WEI (-0.09 d/year). The results also showed that the variability of sow performance increased over time for TNB, NBA and SWL and decreased for WEI. Similarly, the variability of birth weight and of average daily gain from birth to 21d of age was higher in D8 than in D7 piglets. This higher variability might be an indicator of a lower sow and piglet robustness.

**Keywords:** efficiency, reproduction and maternal abilities, selection, variability

## Introduction

Selection has been implemented in pig populations for several decades. Initially, the breeding goal was to increase growth, feed efficiency and carcass lean content. Since the early 1990's, litter size has become a major component of the breeding goal in French Large White (LW) and Landrace (LR) breeds, leading to large genetic gains on litter size (Tribout *et al.*, 2003; Guéry *et al.*, 2009). This improvement may unfortunately be accompanied by detrimental effects on other economically important traits. For instance, an increase in piglet mortality was shown in French LW by Tribout *et al.* (2003) and Canario *et al.* (2007). Animal robustness, defined as the ability to maintain a good level of performance over a wide range of environments (Knap, 2005) could also unfavourably be affected by selection (Phocas *et al.*, 2014).

The adverse effects of selection are difficult to detect, as only a limited number of traits are routinely recorded in breeding programmes. The use of frozen semen is a simple and powerful way of measuring genetic trends for a large number of traits (Smith, 1977). This approach has been used in French LW population to estimate genetic trends from 1977 to 1998. Tribout *et al.* (2010) estimated genetic trends for growth and carcass traits. Preliminary results for reproduction traits were presented by Tribout *et al.* (2003).

The objective of this study is to analyse the full set of data of the experiment described by Tribout *et al.* (2003; 2010) and investigate the effects of selection on the variability of sow and piglet performance as a potential indicator of their robustness.

## Materials and Methods

### Animals

French large white sows were inseminated in the INRA GENESI experimental unit (17700 Surgères) with either stored frozen semen from 17 LW boars born in 1977 (G7) or with semen from 23 LW boars

born in 1998 (G8). Thirty and 33 litters, respectively, were produced from G7 and G8 boars. After weaning, half of piglets from each litter and sex were transferred to the INRA experimental herd of Bourges (18520 Avord). Fifteen males from each group, as well as 74 G7 and 89 G8 females were chosen at random to produce a second generation by within group matings. Pigs from this second generation will be noted as D7 and D8.

D7 and D8 sows were kept for 6 litters. They were managed under a batch farrowing system, with three weeks intervals between successive batches. They were inseminated twice when oestrus was detected. Farrowing was induced with prostaglandin on the 113<sup>th</sup> day of gestation. The day of farrowing, total number born (TNB), number of stillbirths (SB) and number born alive (NBA) were recorded for each litter. All piglets born were individually weighed within 24 hours after farrowing.

In order to disentangle sow and litter effects on piglet growth, cross fostering was used on a large scale in order to let sows from each group nurse 50% of their own piglets and 50% piglets from the other genetic group, and litter sizes were standardised (D7 and D8 sows nursed 7 and 12 piglets/litter, respectively). Sows were fed ad-libitum from about d 5 of lactation and their daily feed consumption was recorded. Conversely, no creep feed was distributed to piglets before 21 days (d) of age in order to accurately estimate sow milk production. The piglets were weaned at four weeks of age (28 d on average). Piglets were individually weighed at 21 d of age and at weaning.

Sow traits analysed included TNB, NBA, SB, litter weight at birth (LWB), litter growth weight from birth to 21 d of age (LGR21d), weaning to oestrus interval (WOI), total (SWL<sub>t</sub>) and net (SWL<sub>n</sub>) sow weight loss during lactation - SWL<sub>t</sub> = sow weight at farrowing (SWF) minus sow weight at weaning (SWW); SWL<sub>n</sub> = SWF - SWW - (0.3 + 1.329 LWB) (Dourmad *et al.*, 1997). Sow longevity (LONG) was computed as lifespan from birth to last weaning. Piglet traits included individual weight at birth (IWB) and at 21 d of age (IW21d), average daily gain from birth to 21 d of age (ADG21d) and to from birth to weaning (ADGW).

### Statistical Analysis

Traits were analysed using mixed linear models with the SAS Mixed procedure. The model for sow traits included genetic group, herd, farrowing batch within herd and parity as fixed effects, sow and litter sire as random effects. The model for piglet traits included the same effects as for sow traits, plus litter genetic group and a sex effect. Interactions between fixed effects were tested in preliminary analyses and kept in final models when significant.

The homogeneity of residual variances across genetic groups (D7 vs D8) was also tested using SAS MIXED procedure. When variances significantly differed, the final analyses were performed using a model accounting for different variances in D7 and D8 groups. The estimated annual genetic trend ( $\Delta Ga$ ) and its standard error (SE) for each trait were computed using the following formulae (Smith, 1977):  $\Delta Ga = (2 * (\mu_{D8} - \mu_{D7})) / 21 \pm (2 * SE_{D8-D7}) / 21$ , Where  $\mu_{D8}$  and  $\mu_{D7}$  are estimates of genetic group effects and  $SE_{D8-D7}$  the standard error of the contrast between D8 and D7.

### Results and Discussion

The annual genetic trend and group least squares means for sow traits are given in Table 1. D8 sows had larger litter sizes and weights than D7 sows; gains from 1977 to 1998 were  $2.48 \pm 0.96$  and  $1.89 \pm 0.95$  piglets for TNB and NBA, respectively, and  $4.78 \pm 1.21$  kg for LWB. The lower genetic trend for NBA as compared to TNB resulted from an increase in the number of stillbirths (SB: +0.68 piglet,  $p < 0.09$ ). The percentage of stillbirths was not globally different between groups, but was higher for D8 in the latter parities.

The large increase in LWB ( $>1$  kg every five years -  $P < 0.0007$ ) indicated an improved ability of D8 sows to provide nutrients to their foetuses during gestation, i.e. an improved “uterine capacity”. Conversely, with a similar number of piglets nursed, LGR21d was lower in D8 as compared to D7 sows, which indicates a decreased milk production of D8 as compared to D7 sows. SWL<sub>t</sub> was higher in D8 as compared to D7 sows (Table 1), corresponding to a genetic trend of 11.2 kg after 21 years of selection. Yet, as shown by the lack of group difference for SWL<sub>n</sub>, this higher loss was entirely due to the increase in piglet and embryonic tissue weights. The interval from weaning to first oestrus was more than one day shorter in D8 sows ( $p < 0.01$ ), as compared to D7 sows.

Table 1. Least squares means and annual genetic trends for sow performance

Trait <sup>1</sup>	Observation		Mean performance		$\Delta Ga \pm SE$	Pr >  t  for H0 : $\Delta Ga = 0$
	D7	D8	D7	D8		
TNB	252	283	11.59 ± 0.52	12.83 ± 0.45	0.12 ± 0.06	0.009
NBA	252	283	10.40 ± 0.52	11.29 ± 0.44	0.09 ± 0.06	0.050
SB	252	283	1.18 ± 0.24	1.52 ± 0.21	0.03 ± 0.03	0.09
LWB (kg)	220	237	15.81 ± 0.70	18.20 ± 0.62	0.22 ± 0.08	0.0007
LGR21d (kg)	185	196	2.64 ± 0.09	2.41 ± 0.10	-0.02 ± 0.13	0.06
SWL <sub>t</sub> (kg)	228	251	32.7 ± 2.1	38.3 ± 2.2	0.52 ± 0.28	0.04
SWL <sub>n</sub> (kg)	228	251	15.2 ± 2.6	14.2 ± 2.4	-0.09 ± 0.33	0.7
WOI (day)	107	109	5.6 ± 0.3	4.4 ± 0.3	-0.10 ± 0.04	0.001
LONG (day)	252	283	354 ± 34.5	261 ± 39.3	-8.8 ± 3.87	0.02

<sup>1</sup>TNB = total number born; NBA = number born alive; SB = number of stillbirths; LWB = litter weight at birth; LGR21d: litter growth rate from 0 to 21 days of age; SWL<sub>t</sub> = total sow weight loss from farrowing to weaning; SWL<sub>n</sub> = net sow weight loss from farrowing to weaning (see text); WOI = weaning to oestrus interval; LONG = sow longevity.

Table 2 shows the effects of both sow and piglet genetic group on individual piglet weights and growth rate. Though born in larger litters, D8 piglets tended to be heavier at birth than D7 piglets (+66g; P=0.07). Least squares for IWB<sub>cf</sub> clearly showed that the heaviest piglets were cross fostered in both groups, with a difference of 48 g and 74 g over non cross fostered piglets in D7 and D8 groups, respectively). Conversely, the average IWB<sub>cf</sub> of piglets nursed by D7 and D8 sows were similar (respectively, 1482 g and 1469 g).

In accordance with results on LGR21d, piglets nursed by D7 sows had a larger ADG21d than those nursed by D8 sows (+12 g/d), with a larger difference for D7 as compared to D8 piglets (+17 g/d vs +7 g/d). As a result, piglets nursed by D7 sows were heavier at 21d than those raised by D8 sows (+0.36 kg and +0.18 kg, respectively, for D7 and D8 piglets). Globally, the effect of piglet genetic group was limited (D8-D7 equals +2 g/d and -0.12 kg, respectively, for ADG21d and IW21d). Adjusting the 2 traits for IWB has a limited impact on the results. Estimated genetic trends were non significant at the piglet level and negative (estimates of  $\Delta Ga$  for ADG21d<sub>aj</sub> and IW21d<sub>aj</sub> were -1.3 g/d and -14 g, respectively).

Table 2. Least squares means for piglet performance

Traits <sup>1</sup>	Mean performance				P-value
	D7		D8		
Sows	D7		D8		
Piglets	D7	D8	D7	D8	
IWB (g)	1441 ± 33		1507 ± 28		0.07
IWBcf (g) *	1416 ± 30 <sup>a</sup>	1548 ± 31 <sup>c</sup>	1464 ± 29 <sup>ab</sup>	1474 ± 25 <sup>b</sup>	0.0001
ADG21d (g/d)	235 ± 6 <sup>a</sup>	232 ± 7 <sup>a</sup>	218 ± 6 <sup>b</sup>	225 ± 5 <sup>ab</sup>	0.01
ADG21d <sub>aj</sub> (g/d)	237 ± 6 <sup>a</sup>	231 ± 7 <sup>a</sup>	219 ± 6 <sup>b</sup>	222 ± 5 <sup>ab</sup>	0.02
IW21d (g) *	6.40 ± 0.16 <sup>a</sup>	6.43 ± 0.16 <sup>a</sup>	6.04 ± 0.14 <sup>b</sup>	6.25 ± 0.13 <sup>ab</sup>	0.06
IW21d <sub>aj</sub> (g) *	6.49 ± 0.16 <sup>a</sup>	6.38 ± 0.16 <sup>a</sup>	6.11 ± 0.14 <sup>b</sup>	6.15 ± 0.13 <sup>ab</sup>	0.08

<sup>1</sup> IWB = piglet individual weight at birth; IWBcf = individual weight at birth after cross fostering; IW21d = individual weight at 21 days old; ADG21d = average daily gain until 21 days old; \* different superscript in the same line is significant different.

Within group residual standard deviations (RSD) for each trait are given in Table 3. RSD was significantly larger in D8 than in D7 sows for NBA and WOI, as well as for piglet traits. The increased RSD of D sow indicates that their performance is more variable during their productive life. This result is in agreement with Johnson et al. (1999), who also showed an increased variability of performance at birth as a result of selection for litter size. This higher variability can be considered as detrimental for both physiological and management reasons. This heterogeneity may partly explain the significantly higher within-litter variability of D8 piglets at birth. A higher variability is also observed for ADG21d and IW21d, but it is more difficult to interpret, as piglets were not randomly allocated to their nurse genetic group.

Table 3. Standard deviation of residual variance of sow traits

Trait <sup>1</sup>	Standard deviation		P-value
	D7 <sup>2</sup>	D8 <sup>2</sup>	
TNB (piglet)	2.32 ± 0.16	2.65 ± 0.15	0.13
NBA (piglet)	2.14 ± 0.15	2.60 ± 0.14	0.03
LW21d (kg)	7.68 ± 0.70	8.62 ± 0.65	0.32
WOI (day)	1.79 ± 0.28	1.09 ± 0.29	0.0001
IWB (g)	279 ± 10	306 ± 9	0.002
IW21d (g)	1037 ± 58	1158 ± 51	0.003
ADG21d (g)	43 ± 2	49 ± 2	0.001
	D7 <sup>3</sup>	D8 <sup>3</sup>	
IWB (g)	269 ± 10	305 ± 9	0.0001
IW21d (g)	1016 ± 58	1136 ± 51	0.003
ADG21d (g)	44 ± 2	49 ± 2	0.002

<sup>1</sup> TNB = total number born; NBA = number born alive; WOI = weaning to first oestrus interval; IW21d = individual weight at 21 days of age; ADG21d = average daily gain until 21 days of age; <sup>2</sup> genetic group of nurse sows; <sup>3</sup> genetic group of piglets.

## Conclusion

This study shows that selection in French LW breed has resulted in strong improvement of sow performance during gestation. Globally, modern sows are much more productive at birth than old sows. However, selection has not been accompanied by an improvement in sow milking performance, has had detrimental effects on sow longevity and has resulted in more heterogeneous performances during their productive life. These trends could be interpreted as a decrease in sow robustness. Taking into account these traits in future breeding goals in sow dam lines might be desirable.

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# Effect of Caffeine on Morphology of Epididymis Spermatozoa of Bali Bull

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## Abstract

*There is an unexpected loss of breeding animals of high genetic value for any reason, such as loss of libido, reproductive tract injury, or death. This loss can be reduced by harvesting spermatozoa from the epididymis with cryopreservation. However, after cryopreservation, there is still a decline of 50% in sperm post-thaw viability after thawing process. This research was conducted to evaluate the use of caffeine to improve cryopreservation quality of Bali bull's spermatozoa. Selection for sperm quality was based on sperm morphology. Cauda epididymis was obtained from slaughterhouse and the semen was collected and examined with caffeine after cryopreservation. The spermatozoa were stained with Acredine Orange by microscopy laser-scanning confocal microscope (BioRad MRC-1024). The result demonstrated that control have high value of normal sperm cell than those treated with caffeine.*

*Key words: cauda epididymis, abnormality, caffeine*

## Introduction

Bali cattle are widely spread in Eastern Indonesia because they have good adaptability and have a fairly high carcass that reaches 57%. Bali cattle were originated from a wild ox of java that had been domesticated in Indonesia. Well-maintained Bali cattle will show a good performance, so the male Bali cattle that are genetically well will show a color change from brown brick to a black color. In order not to lose these genetic resources sperm cryopreservation of Bali cattle should be conducted.

Cryopreservation is one way of handling the sperm that is long lasting and can be used at any time as needed. However, cryopreservation of sperm is often experienced problems such as loss of viability of sperm that can occur because of freezing process. Loss of post thawing motility can reach 50% (Lemma, 2011), and therefore it is needed for the addition of additives before artificial insemination (AI). Caffeine is one of the ingredients that can be added in post-thawing sperm.

Caffeine is an alkaloid found in plants formed of white powder with the mechanism of action to inhibit the activity of nucleotide phosphodiesterase. Nucleotide phosphodiesterase inhibited the production of cAMP and cAMP levels are relatively low due to the activity of nucleotide phosphodiesterase. The addition of caffeine can suppress the activity of nucleotide phosphodiesterase that eventually increases cAMP levels (Hasbi et al, 2011). Increased nucleotide phosphodiesterase will increase motility, thus further improving sperm metabolism when in certain circumstances it is possible to cause damage or abnormal spermatozoa.

## Materials and Methods

### Cryopreservation sperm

Collection of spermatozoa from cauda epididymis by making an incision in the cauda epididymis then put in a test tube medium Tris until 3 minutes, and then continued the process of cryopreservation. Add egg yolks 20% in medium tris, ekulibrasi at a temperature of 4 degrees celsius for 2 hours and then to add to the straw-sized 0,25ml. Take place it in a styrofoam plate in liquid nitrogen vapor for 20 minutes and then put in a storage container for the liquid nitrogen.

### Morphological examination of spermatozoa

The frozen semen was thawed in a water bath at 38°C for 30 seconds, centrifuged at 1800 rpm for 5 second before being used in each experiment. Sperm head morphology was studied in thin smears prepared, fixation 2 h in acetic acid: methanol = 1:3) and stained with Acredine Orange (AO). Two hundred spermatozoa were counted in each smear at a magnification of 1200x in a light microscope laser-scanning

confocal microscope (BioRad MRC-1024). Two hundred spermatozoa were counted in each preparation and the abnormalities (acrosomes, nuclear pouches, proximal and distal cytoplasmic droplets, midpiece and abnormal tails) were classified according to a system developed by Bane (1961). The number of spermatozoa showing each class of abnormality was expressed as a percentage of the counted spermatozoa.

### Statistical analysis

The differences between concentrations were compared and results were expressed as mean  $\pm$  Sd. Analysis of variance (ANOVA) using the SPSS software version 18 with Tukey test was performed to verify statistical significance. The p-values of  $<0.05$  was considered as statistically significant.

### Result and Discussion

Morphological evaluation of spermatozoa is very important because it indicates the spermatogenesis process running normally during sperm maturation in the epididymis that can be seen from the abnormality. Morphological parameters with low value would be detrimental because it can decrease the fertility of sperm after insemination.

Epididymis is a place for maturation of spermatozoa. Spermatozoa were left testes still have a morphology that is not perfect and sperm membrane integrity is still weak, so that possible damage to spermatozoa possible higher than the ejaculation of sperm membrane integrity. Cryopreservation process on epididymal spermatozoa is also very possible spermatozoa is damaged or abnormality, due to changes in temperature (cold shock) when the cryopreservation. Table 1 will be obvious epididymal spermatozoa damage as a result of incomplete morphology of spermatozoa leave the testes and cryopreservation process.

Abnormality	Level of Caffeine (mg/ml in %)			
	0	2	4	6
Normal	45.30 (1.97 $\pm$ 0.42) <sup>a</sup>	44.32(1.70 $\pm$ 0.77) <sup>ab</sup>	43.73 (1.37 $\pm$ 0.63) <sup>b</sup>	43.20 (1.44 $\pm$ 0.68) <sup>b</sup>
Pear Shape	1.68 (0.21 $\pm$ 0.00) <sup>a</sup>	1.41(0.47 $\pm$ 0.21) <sup>a</sup>	1.09 (0.55 $\pm$ 0.26) <sup>a</sup>	2.68 (0.45 $\pm$ 0.36) <sup>a</sup>
Narrow	2.74 (0.74 $\pm$ 0.20) <sup>a</sup>	2.11(0.53 $\pm$ 0.36) <sup>a</sup>	1.8 (0.36 $\pm$ 0.00) <sup>a</sup>	0.90 (0.30 $\pm$ 0.00) <sup>a</sup>
Narrow at the base	1.47 (0.25 $\pm$ 0.09) <sup>a</sup>	0.7(0.35 $\pm$ 0.00) <sup>a</sup>	1.44 (0.36 $\pm$ 0.00) <sup>a</sup>	0.90 (0.30 $\pm$ 0.00) <sup>a</sup>
Abnormal contour	0.42 (0.21 $\pm$ 0.00)	-	-	0.60 (0.30 $\pm$ 0.00)
Round underdeveloped	0.84 (0.21 $\pm$ 0.00)	-	1.08 (0.36 $\pm$ 0.00)	2.08 (0.42 $\pm$ 0.16)
Underdeveloped	7.63 (0.51 $\pm$ 0.28) <sup>a</sup>	9.17(0.57 $\pm$ 0.34) <sup>ab</sup>	11.22 (0.51 $\pm$ 0.27) <sup>ab</sup>	10.37 (0.69 $\pm$ 0.38) <sup>b</sup>
Macrocephalic	1.05 (0.35 $\pm$ 0.12) <sup>a</sup>	2.45 (0.35 $\pm$ 0.00) <sup>a</sup>	3.96 (0.36 $\pm$ 0.00) <sup>a</sup>	1.79 (0.36 $\pm$ 0.13) <sup>a</sup>
Microcephalic	4.63 (0.31 $\pm$ 0.14) <sup>a</sup>	5.29 (0.59 $\pm$ 0.47) <sup>a</sup>	5.08 (0.64 $\pm$ 0.43) <sup>a</sup>	3.84 (0.64 $\pm$ 0.29) <sup>a</sup>
Decapitated Head	-	-	-	1.49 (0.37 $\pm$ 0.15)
Diadem	-	2.11 (0.42 $\pm$ 0.16)	1.44 (0.36 $\pm$ 0.00)	-
Simple bend tail	24.81 (1.18 $\pm$ 0.61) <sup>a</sup>	27.29 (1.24 $\pm$ 0.51) <sup>a</sup>	26.93 (0.93 $\pm$ 0.57) <sup>a</sup>	29.62 (1.02 $\pm$ 0.53) <sup>a</sup>
Double tail	1.48 (0.30 $\pm$ 0.19)	-	-	-
Coiled tail	1.27 (0.42 $\pm$ 0.22)	-	-	-
Droplet Cytoplasmic distal	1.90 (0.38 $\pm$ 0.18)	2.46 (0.41 $\pm$ 0.15)	-	-
Head sperm	2.73 (0.23 $\pm$ 0.06) <sup>a</sup>	1.41 (0.47 $\pm$ 0.21) <sup>b</sup>	0.72 (0.36 $\pm$ 0.00) <sup>ab</sup>	0.90 (0.30 $\pm$ 0.00) <sup>ab</sup>
Tail sperm	2.10 (0.26 $\pm$ 0.10) <sup>a</sup>	1.06 (0.53 $\pm$ 0.25) <sup>a</sup>	1.08 (0.36 $\pm$ 0.00) <sup>a</sup>	1.49 (0.37 $\pm$ 0.15) <sup>a</sup>
Total abnormality	54.70 (0.48 $\pm$ 0.47) <sup>a</sup>	55.68 (0.69 $\pm$ 0.49) <sup>b</sup>	56.27 (0.60 $\pm$ 0.43) <sup>ab</sup>	56.80 (0.67 $\pm$ 0.47) <sup>b</sup>

Different superscripts in the same row means significantly different ( $P<0.05$ )

The results showed that a large degree of abnormality in the simple bend tail (24.81%, 27.29%, 26.93%, and 29.62%) respectively and underdeveloped (7.63%, 9.17%, 11.22%, and 10.37%), respectively with the 0, 2, 4, and 6 mg% caffeine. Abnormality in the tail is due to the post-collection process, whereas underdeveloped abnormalities caused by genetic factors, or during the process of spermatogenesis. Post-collection handling and sperm freezing process causes many spermatozoa abnormalities in the tail. Epididymis spermatozoa have membrane stability are more susceptible to cold shock and osmotic pressure (Hewitt et al. 2001). Underdeveloped abnormality or often called teratoid occurs because of degeneration of primordial cell in the seminiferous tubules (Barth and Oko 1989).



# Comparisson of Anglo Nubian X Etawah Grade and Saanen X Etawah Grade Goats for Some Reproductive Traits

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## Abstract

Demands of goat milk is increasing, however the production of local goat milk is still low. To increase goat milk production is through crossbreeding of local goats with exotic goat breeds. A study was done to comparing some reproductive traits of Anglo Nubian (AN) x Etawah grade (PE) goats and Saanen (SA) x Etawah Grade goats. There were 32 does at first kidding period that consisted of 14 AN x PE does and 18 SA x PE does. All does were managed in the same feeding system at Dairy Goat Unit of Indonesian Research Institute for Animal production. Data were analyzed using linear model from SAS program. Results had showed that numbers of services to conception, gestation length, age of first kidding AN x PE and SA x PE were not different ( $P > 0.05$ ). However, post-partu estrus and litter-size were different between the genotypes ( $P < 0.01$ ). It can be concluded that AN x PE crossbred showed better reproduction performance. This study might be used as early information for recommendation in increased goat milk production using Anglo Nubian breed.

Keywords: Anglo Nubian, crossbreeding, Etawah Grade, reproduction, Saanen

## Introduction

Goat milk has been favorable for consumption like cow milk, even for infant. It has been well-known that goat milk has several medicinal values as therapeutic virtues for dietetic and ulcers problems or people allergic to cow milk and inflammatory diseases (Kumar *et al.*, 2012; Asresie and Adugna, 2014). Therefore, people are willing to pay higher for goat milk than cow milk, in many country (Kosgey *et al.*, 2013).

Etawah grade, a local goat breed, is one of the dairy goat breed in Indonesia. They have been well-known for their adaptability in harsh environment thus smallholder farmers like to raise them. However, their milk production is still low ranged 0.2-1.2 liter/head/day (Sutama *et al.*, 2014; Praharani, 2014a).

Increasing goat milk is through genetic improvement either by selection or crossbreeding. Many crossbreeding program in dairy goat has been done to increase goat productivity in producing milk (Assan, 2013; Norberg *et al.*, 2014). Etawah grade has been crossbred to Saanen since many year ago, resulted to increased milk production 0,8-1,2 liter/head/day (Sutama *et al.*, 2011).

Indonesian Research Institute for Animal Production has done a crossbreeding program using Anglo Nubian bucks mated to Etawah Grade does since 2012. First crossbred kids were born in 2013 and have been observed their productivity. Praharani (2014b) and Praharani *et al.* (2014b) reported that growth rate from birth to puberty of F1 AN x PE was higher than Ettawah Grade.

Reproduction performance of females such as fertility, gestation length, age of first kidding, kidding interval are important factors affected production efficiency (Bhowmik *et al.*, 2014). The profitability of goat production depends primarily on the efficiency of female productivity. Improving female's reproductive performance by selection is difficult due to its low heritability (García-Peniche *et al.*, 2012) indicating that the possibility for genetic improvement through selection is limited, therefore, crossbreeding can be used to quickly improve reproductive performance via its resultant yield of heterosis (Hassan *et al.*, 2007).

The objective of this study was to compare some reproductive traits of Anglo Nubian (AN) x Etawah grade (PE) goats and Saanen (SA) x Etawah Grade goats. This study might be used as early information for recommendation in increased goat milk using Anglo Nubian breed.

## Materials and Methods

The study was carried out at Dairy Goat Unit, Indonesian Research Institute for Animal Production, in Bogor District located about 300 m above sea level.

There was 32 does used in this study consisting of 14 F1 AN x PE does and 18 SA x PE does. All animals were grouped based on genotype (AN x PE and SA x PE) and body weights (25-30 kg) which were

4-5 animals/group. All does aged between 8 and 10 months. Does were mated naturally when estrus with each bucks breed. After kidding, does and kids were separated.

The animals were raised in the same management system. They were fed 500-600 gram/doe/day of concentrate mixture containing 17% crude protein 70% TDN. About 3-4 kg of King Grass and 200-300 gram of legumes mixture containing *Gliricidae sp* (gamal), *Caliandra*, *Leucaena leucocephala* (lamtoro) were available for animals in the pen. Water were *ad libitum*.

Parameters observed were numbers of services to conception, gestation length, litter size, age of first kidding, and post-partus estrus. Numbers of services to conception was defined as how many doe had been mated until they were pregnant. Gestation length was measured from mating until kidding. Post-partus estrus was the day when the first estrus came after kidding. Post-partus estrus was observed when the doelings showed overt estrus signs by waggling its tail, bleating, mounting others and/or allowed the bucks or other goats to mount her (Hafez, 2000)

All data were analyzed using general linear model (SAS, 2003). P-Diff was used to test significantly the genotypes (AN x PE and SA x PE).

## Results and Discussion

### Numbers of Services to Conception (NSC)

The descriptive statistics for the traits evaluated are shown in Table 1. The overall means of numbers of services to conception was 1.38, with ranged between 1-3 and high variation of 48.66%. This results was in agreement of some literature stated that numbers of services to conception of goats ranged between 1-2.75 (Sutama, 2009; Abdullah *et al.*, 2012; Parasmawati *et al.*, 2013; Abdalla *et al.*, 2015). Abdullah *et al.* (2012) reported numbers of services to conception in Shami Goat in Malaysia was 1.1-1.2. The PE females has low pregnancy rate (60 – 73%) when mating in the first estrous usually resulted in low pregnancy rate reported by Sutama (2009) that mean numbers of services to conception > 1.4. Abdalla *et al.* (2015) stated that number of services to conception in Saanen raised in Sudan was  $1.53 \pm 0.12$ .

Table 1. Descriptive statistics for the reproductive traits analyzed

Traits	N	Means	Min.	Max.	Standard deviation	Coefficient variation
Numbers mating to pregnant	32	1.38	1	3	0.62	48.66
Gestation length, days	32	150.83	148	158	2.71	5.91
Age of First kidding, days	32	467.22	397	503	41.12	1.79
Litter size	32	1.54	1	2	0.49	27.04
Post-partus estrus, days	32	38.93	31	51	5.08	38.93

The least square means (LSMeans), standard error and P-value from different genotypes between AN x PE and SA x PE are shown in Table 2. Genotype was not affect numbers of services to conception ( $P > 0.05$ ) in this study. Numbers of services to conception were  $1.31 \pm 0.18$  and  $1.44 \pm 0.15$  for AN x PE and SA x PE, respectively. This results were similar to some findings that numbers of services to conception were not affected by genotypes or breed of goats (Parasmawati *et al.*, 2013). The PE x Boer goats showed numbers of services to conception  $2.75 \pm 1.35$  (Parasmawati *et al.*, 2013).

Table 2. Reproductive traits by genotypes

Traits	P-value	LSMeans $\pm$ standard error	
		AN x PE (N=14)	SA x PE (N=18)
Numbers of services to conception	0.5821	$1.31 \pm 0.18$	$1.44 \pm 0.15$
Gestation length, days	0.2767	$150.52 \pm 0.56$	$151.75 \pm 0.95$
Litter size	0.0109	$1.73^a \pm 0.10$	$1.32^b \pm 0.09$
Age of First kidding, days	0.6272	$469.39 \pm 8.68$	$461.00 \pm 14.72$
Post-partus estrus, days	0.0035	$37.43^a \pm 0.92$	$43.25^b \pm 1.51$

### Gestation Length

The overall means of gestation length was 150.83 days with ranged between 148-158 days and low variation of 5.91% (Table 1). This results was in agreement of some literature stated that gestation length

of goats ranged between 144-156 days (Sutama, 2009; Parasmawati *et al.*, 2013; Patel and Pandey, 2013; Bhowmik *et al.*, 2014). Gestation period of Jamunapari, Black Bengal and crossbred goats were  $151.71 \pm 8.19$ ,  $146.72 \pm 7.61$  and  $147.85 \pm 7.74$  days, respectively (Bhowmik *et al.*, 2014). Patel and Pandey (2013) stated that gestation length of Mehsana goat from India was  $148.97 \pm 0.28$  days.

Genotype was not affect gestation length ( $P>0.05$ ) in this study. Gestation length were  $150.52 \pm 0.56$  days and  $151.75 \pm 0.95$  days for AN x PE and SA x PE, respectively (Table 2). This results were similar to some findings that gestation length were not affected by genotypes or breed of goats (Parasmawati *et al.*, 2013). The PE x Boer does had 152.41 days for gestation length (Parasmawati *et al.*, 2013).

Gestation length of PE goats was between 144-156 days summarized by Sutama (2009), Saanen 150 days (Sabil *et al.*, 2011) in Sudan condition. Anglo Nubian howed gestation length  $147.1 \pm 0.8$  days in Saudi Arabia (Yagoub *et al.*, 2013) and  $152.3 \pm 5.4$  in Sudan (Hassna *et al.*, 2013).

### Age at First Kidding

The overall means of age of first kidding was 467.22 days with ranged between 397-503 days and 1.79% (Table 1). This results was in agreement of some literature stated that age of first kidding in goats ranged between 321-534 days (Sutama, 2009; Patel and Pandey, 2013; Bhowmik *et al.*, 2014) and 11.9-28.5 months for local goats in Ethiopia summarized by Dereje *et al.* (2015). In Iran, age at first kidding of Jamunapari, Black Bengal and crossbred goats were  $534.00 \pm 24.58$ ,  $368.12 \pm 16.96$  and  $471.25 \pm 21.25$  days, respectively (Bhowmik *et al.*, 2014). Mehsana goat from India was at  $716.52 \pm 19.01$  days (Patel and Pandey, 2013)

Genotype was not affect age of first kidding ( $P>0.05$ ) in this study. Age of first kidding were  $469.39 \pm 8.68$  days and  $461 \pm 14.72$  days for AN x PE and SA x PE, respectively (Table 2). Dereje *et al.* (2015) found there is breed differences in age of first kidding.

Puberty in PE goat summarized by Sutama (2009) was attained at 321-362 days of age, therefore age of first kidding might be at 471-512 days. In the present study, age of first kidding was younger than Sutama (2009) due to different management resulted in different body weight. Abdalla *et al.* (2015) stated that number of services to conception in Saanen raised in Sudan was  $458.11 \pm 11.89$  day. The pooled data of Saanen, Toggenburg, Anglo Nubian and La Mancha in USA dairy goats showed that age of first kidding was 507.97 days (Castañeda-Bustos *et al.*, 2014)

### Litter Size

The overall mean of litter size 1.54, ranged between 1-2 and variation 27.04 % (Table 1). This results was in agreement of some literature stated that litter size of goats ranged between 1-1.7 (Sutama, 2009; Mellado *et al.*, 2011; Parasmawati *et al.* 2013).

Genotype affected litter size ( $P<0.01$ ) in this study. Litter size were  $1.73 \pm 0.10$  and  $1.32 \pm 0.09$  for AN x PE and SA x PE, respectively (Table 2). This results were similar to some findings that Litter size were not affected by genotypes or breed of local goats in Ethiopia (Dereje *et al.*, 2015). Mellado *et al.*, (2011) reported in Mexico Saanen ( $1.34 \pm 0.52$ ) showed lower litter size than Anglo Nubian ( $1.56 \pm 0.5$ ). However Parasmawati *et al.* (2013) found that litter size were no significantly different between genotype.

Litter size of PE goats was 1-1.04 that will increase to 1.3-1.6 as parity increased (Sutama, 2009). While Parasmawati *et al.* (2013) mentioned that PE x Boer goat had litter size  $1.70 \pm 0.86$ . Anglo Nubian in Sudan showed litter size  $1.75 \pm 0.71$  (Hassna *et al.*, 2013). In Sudan condition, litter size of Saanen was 2.5 (Sabil *et al.*, 2011)

### Post-partus Estrus

Post-partus estrus ranged between 31-51 days with average of 38.93 days and variation of 38.93% (Table 1). This result was in agreement of some literature stated that post-partus estrus of goats ranged between 23-74 days (Frietas *et al.*, 2004; Sutama, 2009). Another study showed that the kidding interval of Jamunapari, Black Bengal and crossbred goats were  $224.00 \pm 14.42$ ,  $181.76 \pm 15.81$  and  $199.17 \pm 21.71$  days, respectively, which means that post-partus estrus at 31-74 days (Bhowmik *et al.*, 2014).

Genotype affected post-partus estrus ( $P<0.01$ ) in this study. Post-partus estrus were  $37.43 \pm 0.92$  and  $43.25 \pm 1.51$  for AN x PE and SA x PE, respectively (Table 2). This results were similar to some findings that post-partus estrus were affected by genotypes or breed of goats. Freitas *et al.* (2004) mentioned that significant difference was observed for post-partus estrus between Anglo Nubian and Saanen. Anglo-Nubian goats showed a shorter first estrus ( $23.89 \pm 5.64$  days) when compared to Saanen goats ( $46.20 \pm 9.50$  days) similar to the present study that AN x PE shorter than SA x PE.

A review (Sutama, 2009) mentioned that first estrus after kidding in PE goat showed at 3-5 months because does and kids stayed together until weaning. In this study, post-partus estrus showed faster than Sutama (2009), because does and kids were separated after kidding. In Saudi Arabia, Anglo Nubian showed first estrus after kidding at  $51.0 \pm 4.5$  days (Yagoub *et al.*, 2013), and in Sudan  $57.8 \pm 21.5$  days (Hassna *et al.*, 2013).

## Conclusion

Numbers of mating per conception, gestation length, age of first kidding were not different between AN x PE and SA x PE, but post-partus estrus and litter-size. It can be concluded that AN x PE crossbred showed better reproduction rate. From these findings showed performance of AN x PE, a new crossbred does, were satisfactory which indicated their adaptability in the tropics. This study might be used as early information for recommendation in increased goat milk production and quality using Anglo Nubian breed.

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# Service Per Conception In Beef Cattle With Artificial Insemination in Kapuas Basarang District Of Central Kalimantan

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## Abstract

*The purpose of this study was to compare the relative efficiency of the process of reproduction among individuals fertile female cattle, so that breeding have good yield as well as to improve the beef cattle farm in Kapuas Basarang district. The method used in this research is a survey (observations) directly to farmers and data collection was done by interviewing the farmers and participate directly in productive activities of pregnancy examination. Service per conception (S/C) in the A working area is 1.70; the B working area is 3; and the A and B working areas 2.07. The best S/C is in the A working area 1.70. Calving rate (CR) in the A working area is 55.76%; the B working area 23.67%; and the A and B working areas 42.69%. From interviews, the livestock population is getting less due to lack of interest of the public, especially the youth to do business with serious beef cattle farms. Lack of understanding of the cattle breeders in estrous, causing delays in reporting and dropping liquid N<sub>2</sub> and distribution of straw to the inseminator should be done in response to any report so that artificial insemination services by inseminator always fulfilled in every farmer report. The need for additional personnel and additional knowledge inseminator through insemination training, so inseminator has more skill. Need to do counseling and training to farmers about heat detection.*

*Keyword : artificial insemination, service per conception*

## Introduction

### Background

Development of livestock sub-sector as part of the national development received considerable attention from the Government, prior to meeting the food and nutrition through the development of beef cattle business, it is to meet the need of animal protein, especially meat. To achieve these objectives will be pursued business development and application of appropriate technology: (1) Increase the quantity and quality of livestock (2) Maintenance of health, (3) Counseling, (4) Development and provision of infrastructure, (5) Utilization of livestock waste.

In implementing development livestock, attention needs to be given to the development particularly farm people who constitute the bulk of farmers in Indonesia, increasing the role of cooperatives and the participation of private business.

One that can be taken to increase the production of meat and calves or calves is to increase the number of beef cattle ownership and genetic quality of livestock. This can be done by applying Artificial Insemination (AI) in cattle, since semen is used to AI comes from bulls that have good genetic and numbers Service Per Conception on average smaller than natural mating.

AI is a form of reproductive biotechnology in efforts to increase production and beef cattle productivity with the ultimate target to increase the farmers' income in breeders. AI needs to be improved through intensive efforts, continuous with emphasis on aspects of quality improvement and expansion of the range of AI services in the form of Artificial Insemination Services Unit (SPIB) to realize the AI excellence service and socialized.

### Objective

The objective of this study is to compare the relative efficiency of the process of reproduction among individuals female cattle which are fertile, so that is expected to produce a good breed.

## Materials and Methods

### Materials

This study used 89 female cattle and artificial insemination equipments.

### Methods

The method used in this study is a survey (observations) directly to farmers and data collection was done by interviewing the farmers and participate directly in productive activities of pregnancy examination. Pregnancy examination were performed after AI 60 days ago, then that is collecting data on the cows that have been had AI and grouped based on first AI, second AI and third AI. Then grouped pregnancy that resulted from first AI, second AI and third AI. Service per Conception and Calving Rate is obtained.

## Results and Discussion

Based on this study in the field Service Per Conception in beef cattle with artificial insemination on pregnancy value can be seen in A region and B region.

Table 1. Observations data in January-September 2014

Region	AI			Pregnancy		
	I	II	III	I	II	III
A	42	9	1	29	7	1
B	32	5	1	9	5	1
Total A & B	74	14	2	38	12	2

Specification: Region A= Work region of A inseminator; Region B= Work region of B inseminator.

### Service Per Conception

From observations at region A, there were 52 fertile female cattle which inseminate with fertile semen. After pregnancy examination from 52 fertile female cattle, there were 37 pregnant cows is obtained. S/C in region A is 1.70, which means that from a group of females in A region had high fertility level.

Observation in region B, there were 38 fertile female cattle which inseminate with fertile semen. After pregnancy examination from 38 fertile female cattle, there were only 15 pregnant cows is obtained. S/C in Region B is 3, which means that the level of fertility of a group of females in this region is very low.

While the observations resulted in region A and B, there were 89 fertile female cattle population which produce 52 pregnant cattle and S/C value is 2.0. but fertility rate in region B is less than in region A.

Based on the results of observational data from January to September 2014 over the working region A look at the value of S/C was higher than region B. In accordance with Feradis (2010) who argued the value of S/C the normal range is between 1.6 to 2.0. In region A also shows the normal value of 1.70 which means that the lower the value, the higher the fertility of the females in the group. And conversely the higher the value S/C, the lower the value of a group of female fertility.

If we compared it with the data S/C in 2011 of Veterinary Office Kapuas District found that the value of S/C of 2.8 are the highest in the region A. While the S/C in Region B is 6. In 2011 and 2012 S/C in region A is 1.17 and region B is 3.

When viewed from S/C in 2012 and 2013 fertility group of female cattle tend to normal. However, from the cattle population data from year to year become decreased. From the interviewed this is due to lack of interest of the public, especially the youth to do business with serious beef cattle farms. So we could not based on the S/C data from the previous year. This may be caused by four things: 1). S/C depends on proper estrus detection of livestock farmers, from interviews found still a lack of understanding of the cattle in heat or estrus, so there is a delay in reporting; 2). Inseminator readiness in terms of the provision of straw, and the availability of straw used at any time; 3). Need to pay attention to the liquid N<sub>2</sub> used in the container for the storage of straw, a refill to depleted liquid N<sub>2</sub> approximately a month usage; and 4) inseminator skills.

### Calving Rate

The study resulted that calving rate (CR) in region A is 55.76%, in region B is 23.67%, and the region A and B 42.69%. It appears that the value of calving rate in region A is higher than in region B. It figures

that CR is still low and needs to added the knowledge of estrus detection, good AI, improve the personnel skill of AI, and rapid respond of inseminator.

The value of the CR depends on inseminator efficiency job, male cattle fertility, female cattle fertility when insemination and the ability to received fetus in the womb until the time of birth (Feradis, 2010).

## Conclusion

S/C in beef cattle with AI in Kapuas Basarang District is 2.0 that is still in normal range, but the CR value is still low and needs to added the knowledge of estrus detection, good AI, improve the personnel skill of AI, and rapid respond of inseminator.

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# Association of GH|*MspI* and GHRH|*HaeIII* Genes with Milk Components of Holstein-Friesian (HF) Cows under Small Farmers in Lembang, West Java

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## Abstract

Growth hormone (GH) and Growth Hormone Releasing Hormone (GHRH) genes control tissue growth, reproduction and lactation in dairy cattle. This study aimed at evaluating genetic polymorphism and association of Growth Hormone (GH|*MspI*) and Growth Hormone Releasing Hormone (GHRH|*HaeIII*) genes to daily milk components of HF cows raised by small dairy farmers from two locations (Loc-1 = 60hds and Loc-2 = 58 hds) in Lembang District, West Java. Gene variants were genotyped using PCR-RFLP method. Study of association of variant genotypes to individual milk component (fat, SNF, protein and DM) was performed using a linear model with the fixed effects of lactation length (months), lactation period and those two growth genes. Genotyping GH|*MspI* gene at Loc-1 and Loc-2 resulted (+) allele (0.83, 0.41) and (-) allele (0.17; 0.59); whereas for GHRH|*HaeIII* gene in respective locations resulted A allele (0.29; 0.29) and B allele (0.71; 0.71). For Loc-1, GHRH|*HaeIII* gene had a significant effect ( $P < 0.05$ ) on SNF content, with AA genotype cows producing milk with a higher SNF (8.43%) than BB and AB cows (7.74 %; 7.61 %). For Loc-2, GH|*MspI* gene had a significant effect ( $P < 0.05$ ) on fat content with (+/+) cows produced fatter milk (3.97%) than (+/-) and (-/-) cows (3.05%; 3.39%). The conclusion was that there was a fairly good control of GH|*MspI* and GHRH|*HaeIII* genes on milk components in HF cows. Keywords: association, genotype, growth gene, milk component

## Introduction

Milk is a source of highly nutritious animal food required by various levels of society. Milk production and milk quality of domestic dairy cattle should be increased, thereby increasing the selling price of fresh milk. Holstein Friesian (HF) breed is a main producer of national fresh milk. Genetic improvement in milk production and quality therefore needs to be warranted to this HF cattle. Selection to increase milk production and milk quality should be supported by molecular selection to give a faster progress.

Polymerase Chain Reaction-Restriction Fragment Length Polymorphism (PCR-RFLP) technique can be used to develop molecular-based selection. This technique can detect DNA variants of genes related to economic traits in animal, such as milk production and quality of dairy cattle (Dybus, 2002) and growth and carcass quality of beef cattle (Beauchemin *et al.*, 2006). Growth Hormone (GH) gene and Growth Hormone Releasing Hormone (GHRH) gene or somatotrin in gene have crucial roles in controlling bone and muscle tissue growth, mammary glands, lactation and reproduction traits (Akers, 2006). Both genes can be used in Gene Assisted Selection (GAS) programs for molecular selection of lactation traits.

This study aimed at identifying polymorphisms of GH|*MspI* and GHRH|*HaeIII* genes and at investigating their association with milk components in HF cows kept in small dairy farms in Lembang, West Java.

## Materials and Methods

### Research samples

Dairy cows observed were lactating HF cows for a total number of 118 hds. reared in two villages of Pasar Kamis (Loc.-1) (60 hds.) and Cilumber (Loc.-2) (58 hds.), Lembang District, West Java. These cows were at physiological status of between lactation length of 1-6 mo. and lactation period of 1-5. Milk data were one-day milk test of individual animal and cows were commonly kept by farmer under a semi-intensive management.

## Research methods

Genotyping of blood samples of cattle briefly consisted in DNA extraction, amplification by PCR of DNA fragments of GH and GHRH genes, allele identification by RFLP method by cutting PCR products of DNA fragments of the two genes. Primers used for amplification of GH gene were for forward 5'-CCCCGAGACGGGCAAGAATGC-3' and reverse 5'-TGAGCA GGGGCCGGA ACTCA-3' following Zhou *et al.* (2005), while those for GHRH gene were for forward 5'-TGAAGGCTGCTCTGGATGGT-3' and reverse 5'-TGCTTCCTGATGTCCTGGA TAA-3' following Moody *et al.* (1995). GH gene was cut by Pst1 enzyme, while that for GHRH gene was HaeIII enzyme.

Milk protein content was analyzed by for moltitration, for which pas the amount of NaOH used fortitration of milk sample and qas the amount of NaOH used for blank titration. Analysis was also done for fat, solid non fat (SNF) and dry matter (DM) contents.

## Data analysis

Study of association of genotype variants with individual milk components, consisting of fat, SNF, protein and DM using a linear model (with SAS GLM software) including lactation length (months), lactation period and those two growth genes as fixed effects.

## Results and Discussion

### Genotyping and Frequency of Growth Genes

Extraction of DNA fragment of GH gene produced amplicons along 327 bp at intron 3 and exon 4. This PCR product approached the amplicon length of 329 bpas resulted by Zhou *et al.* (2005). Fragments of GHRH gene produced amplicons along 451 bp, at exon 2, intron 2 and exon 3, similar to the one resulted by Moody *et al.* (1995). Genotyping GH|*MspI* gene resulted in three genotypes, namely (+/+) genotype with two bands (104 and 223 bp), (+/-) genotype with three bands (104, 223 and 327 bp) and (-/-) one with only one band (327 bp) as there was no cutting site. Transition of base mutation of C into T causes of a cutting site by *MspI* restriction enzyme. Genotyping GHRH|*HaeIII* gene produced three genotypes, namely AA genotype with three bands (312, 94 and 45 bp), BB genotype with four bands (194, 118, 94 and 45 bp), and AB genotype with five bands (312, 194, 118, 94 and 45 bp). HaeIII enzyme recognizes a cutting site of GG|CC, due to a base change of alkaline C (Cytosin) to A (Adenine).

Genotyping GH|*MspI* gene at Loc-1 and Loc-2 resulted in (+) allele frequencies of (0.83, 0.41, respectively) and (-) allele (0.1667; 0.5948); whereas for GHRH|*HaeIII* gene in respective locations resulted in A allele frequencies of (0.29 in both locations) and B allele (0.71; 0.71). For all location, frequency of (+) allele (0.62) was higher than that of (-) allele (0.38), while B allele (0.71) was relatively higher than that of A allele (0.29). For Loc.-1, a high frequency of (+/+) cows (0.68) was observed, followed by (+/-) (0.30), and very low frequency of (-/-) (0.02). For Loc.-2, genotype (-/-) cows were dominant (0.47), whereas (+/+) (0.28) and (+/-) (0.26) genotypes had similar genotypic frequencies. Based on the values of the allelic frequencies of all three genotypes of these two genes without exceeding  $\geq 0.99$ , as a limit of monomorphic, so it could be stated that GH|*MspI* and GHRH|*HaeIII* genes of the observed HF from two locations were polymorphic.

Zhou *et al.* (2005) in their study for GH|*MspI* gene of Beijing Holstein cows reported a higher frequency of (+) allele (0.83) than that of (-) allele (0.17). While study for GHRH|*HaeIII* gene of HF cows in Poland by Dybus and Grzesiak (2006) reported frequencies of genotypes of AA (0.05), AB (0.31) and BB (0.63); while those for alleles of A (0.21) and B (0, 79) respectively.

### Milk Components

Results of analysis of milk components providing fat, solid non fat (SNF), protein and dry matter (DM) of the observed HF cows were fairly good. The contents of all of these four milk component sex ceeded minimum limits as specified by ISO3141.1:2011 for fresh milk of dairy cattle in Indonesia. Minimum values specified were 3.0% for fat, 2.8% for proteinand 7.8% for SNF. The exception was for protein content of HF cows in Loc.-1 that was a slightly lower than the SNI provision.

Table 1. Mean of milk component (%) in HF cows based on location

Location	Sample	Fat	SNF	Protein	D. Matter
Loc. 1	60	3.40 ± 0.70 <sup>a</sup>	8.21 ± 0.38 <sup>a</sup>	2.77 ± 0.29 <sup>a</sup>	11.64 ± 0.89 <sup>a</sup>
Loc. 2	58	3.54 ± 0.84 <sup>a</sup>	8.21 ± 0.53 <sup>a</sup>	2.85 ± 0.38 <sup>a</sup>	11.80 ± 1.02 <sup>a</sup>
Both	118	3.47 ± 0.77	8.21 ± 0.46	2.81 ± 0.34	11.72 ± 0.96

Description: Sample in head; same letters in similar column showed no difference ( $P > 0.05$ )

Milk quality of HF cows between the two locations did not differ statistically ( $P > 0.05$ ). This was probably due to nearby locations, so that the ways of feeding, management and climatic conditions were almost similar. The observed HF cows in all location produced milk quality that met minimum standards of SNI.

### Association of Variant Genotypes of Growth Genes on Milk Components

Effect of variant genotypes of *GH|MspI* and *GHRH|HaeII* genes on milk components of HF cows were presented in Table 2. In general *GH|MspI* and *GHRH|HaeII* genes presented unclear effects on most milk components. However, SNF content of HF cows at loc.-1 was influenced by *GHRH|HaeIII* gene. This gene had a significant effect ( $P < 0.05$ ) on SNF, with AA cows producing milk with a higher SNF (8.43%) than that of BB (7.74%) and AB (7.61%) cows. Moreover, fat content of HF cows at loc.-2 was affected by *GH|MspI* gene. This gene had a significant effect ( $P < 0.05$ ) on milk fat content, with (+/+) cows producing fatter milk (3.97%) than (+/-) (3.05%) and (-/-) (3.39%) cows. Furthermore, *GH|MspI* gene consistently had a highly significant ( $P < 0.01$ ) effect on milk yield of HF cows in each location as well as in all locations. (+/+) cows produced a higher daily milk yield than cows with (+/-) and (-/-) genotypes.

Results of this study are consistent with those of Dybus (2002) in Poland, where HF cows with (+/+) genotype at *GH|MspI* gene produced milk with a higher than (+/-) and (-/-) genotypes. Another study reported that AA and AB genotypes at *GHRH|HaeII* gene resulted in a higher fat content than BB genotype (Dybus and Grzesiak, 2006). Based on these results, it can be stated that both *GH|MspI* and *GHRH|HaeII* genes could be considered as candidate genes for GAS on milk yield and milk quality in dairy cows.

Table 2. Effect of variant genotypes of *GH|MspI* and *GHRH|HaeII* genes on milk components (%) and milk yield (liter) in HF cows based on location

Location	Gene	Genotype	Sample	Milk	Fat	SNF	Protein	D. Matter
Loc. 1	<i>GH MspI</i>	(+/+)	41	15.94 <sup>a</sup>	3.282 <sup>a</sup>	8.219 <sup>a</sup>	2.749 <sup>a</sup>	11.542 <sup>a</sup>
		(+/-)	18	12.83 <sup>b</sup>	3.697 <sup>a</sup>	8.207 <sup>a</sup>	2.816 <sup>a</sup>	11.906 <sup>a</sup>
		(-/-)	1	9.00 <sup>b</sup>	3.000 <sup>a</sup>	7.920 <sup>a</sup>	2.980 <sup>a</sup>	10.920 <sup>a</sup>
	<i>GHRH HaeIII</i>	AA	3	12.83 <sup>a</sup>	3.700 <sup>a</sup>	8.433 <sup>b</sup>	2.893 <sup>a</sup>	12.133 <sup>a</sup>
		AB	29	14.95 <sup>a</sup>	3.516 <sup>a</sup>	7.738 <sup>a</sup>	2.756 <sup>a</sup>	11.723 <sup>a</sup>
		BB	28	15.05 <sup>a</sup>	3.252 <sup>a</sup>	7.611 <sup>a</sup>	2.758 <sup>a</sup>	11.503 <sup>a</sup>
Loc. 2	<i>GH MspI</i>	(+/+)	16	13.50 <sup>a</sup>	3.970 <sup>a</sup>	8.287 <sup>a</sup>	2.854 <sup>a</sup>	11.930 <sup>a</sup>
		(+/-)	15	10.73 <sup>b</sup>	3.052 <sup>b</sup>	8.243 <sup>a</sup>	2.781 <sup>a</sup>	11.509 <sup>a</sup>
		(-/-)	27	11.72 <sup>b</sup>	3.388 <sup>a</sup>	8.137 <sup>a</sup>	2.881 <sup>a</sup>	11.892 <sup>a</sup>
	<i>GHRH HaeIII</i>	AA	3	14.50 <sup>a</sup>	2.893 <sup>a</sup>	8.483 <sup>a</sup>	2.750 <sup>a</sup>	11.377 <sup>a</sup>
		AB	28	11.911 <sup>a</sup>	3.664 <sup>a</sup>	8.288 <sup>a</sup>	2.893 <sup>a</sup>	11.960 <sup>a</sup>
		BB	27	11.722 <sup>a</sup>	3.492 <sup>a</sup>	8.090 <sup>a</sup>	2.811 <sup>a</sup>	11.688 <sup>a</sup>
Both	<i>GH MspI</i>	(+/+)	57	15.254 <sup>c</sup>	3.405 <sup>a</sup>	8.238 <sup>a</sup>	2.778 <sup>a</sup>	11.651 <sup>a</sup>
		(+/-)	33	11.879 <sup>a</sup>	3.490 <sup>a</sup>	8.223 <sup>a</sup>	2.800 <sup>a</sup>	11.726 <sup>a</sup>
		(-/-)	28	11.625 <sup>a</sup>	3.586 <sup>a</sup>	8.129 <sup>a</sup>	2.885 <sup>a</sup>	11.857 <sup>a</sup>
	<i>GHRH HaeIII</i>	AA	6	13.667 <sup>a</sup>	3.297 <sup>a</sup>	8.458 <sup>a</sup>	2.822 <sup>a</sup>	11.755 <sup>a</sup>
		AB	57	13.456 <sup>a</sup>	3.589 <sup>a</sup>	8.211 <sup>a</sup>	2.833 <sup>a</sup>	11.840 <sup>a</sup>
		BB	55	13.418 <sup>a</sup>	3.370 <sup>a</sup>	8.177 <sup>a</sup>	2.784 <sup>a</sup>	11.594 <sup>a</sup>

Description: Sample in head; different letters in similar column showed significant difference ( $P < 0.05$ )

## Conclusion

Both GH|*MspI* and GHRH|*HaeII* genes were polymorphic in the observed HF cows. A fairly good indication of control of GH|*MspI* and GHRH|*HaeII* genes on milk components was obtained. Both genes could be considered in a GAS program for molecular selection on milk yield and milk quality in HF dairy cows.

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# Morphological Genetic Distances of Local Buffalo Subpopulations in Pasaman District, West Sumatera Province

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## Abstract

*Estimation of genetic distances through morphology analysis approach for seven body measurements was performed in local swamp buffalo coming from three subpopulations in Pasaman District, West Sumatera Province, Indonesia. Samples were females and males for a total number of 136 heads at ages of 3-5 years. Animals originated from three subdistricts of Lubuk Sikaping (LSi) for 34 hds, Panti (PTi) for 42 hds, and North Rao (NRa) for 60 hds. Genetic variation among those three buffalo subpopulations was predicted by Mahalanobis discriminant function, while genetic distance was estimated by a MEGA program. Male and female buffaloes in both LSi and PTi generally had larger body sizes ( $P < 0.05$ ) compared to those in NRa. Buffalo in NRa had a high degree of similarity (96.67%), while those in LSi and PTi were highly mixing each other. Genetic distance and tree fenogram gave consistent results. Buffaloes between LSi and PTi had a closer genetic distance (0.48838) compared to those of between NRa and LSi (20.96235) as well as NRa and PTi (24.90860). Adjacent geographical condition possibly made an easy access for small farmers to breed buffaloes in LSi and PTi locations.*

*Keywords: discriminant function, genetic distance, morphology, swamp buffalo*

## Introduction

Buffalo in West Sumatra Province is entirely a type of local swamp buffalo, with its population number was for the second largest in Indonesia. Pasaman District is one of important central breeding areas of local swamp buffalo in this Province. Buffalo has been kept for generations by small farmers and has good prospect for future improvement. Nevertheless, by continuously increasing demand of buffalo meat and by converting its habitat to other functions could decrease population and genetic degradation of this animal. Information of the existing animal genetic resources was therefore required in an attempt to conserve and improve genetic quality of local swamp buffalo in this location.

The spread of buffalo population in Pasaman District could be considered as a form of ecological and evolutionary long processes, so morphological differences among subpopulations could be due to genetic effect beside of non-genetic influences. To confirm whether the restriction of area (subdistricts) expressed in morphology was possibly due to an actual genetic difference, so study on the level of genetic information was required (Moritz, 1999). Morphology could be used to determine origin and phylogenetic relationship among different types or breeds of livestock. Genetic distance as an indication of the degree of differences of genes (genome) among (sub)populations could be measured numerically for instance by a Statistical Mahalanobis (D<sub>2</sub>) (Nei, 1987).

This study was aimed to determine genetic variation and phylogenetic relationship through morphometric analysis approach of local swamp buffalo in Pasaman District.

## Materials and Methods

The study was conducted in three subdistricts, namely Lubuk Sikaping (LSi), Panti (PTi), and North Rao (NRa) in Pasaman District, West Sumatra Provinces from July to August, 2009. Figure 1 showed the map of research locations in Pasaman District. A number of local swamp buffalo observed were 136 heads of both male and female at the ages around 3-5 years. Those were respectively from Lubuk Sikaping by 34 heads (M=7; F=27), Panti by 42 heads (M=7; F=35); and North Rao by 60 heads (J=14; B=46).

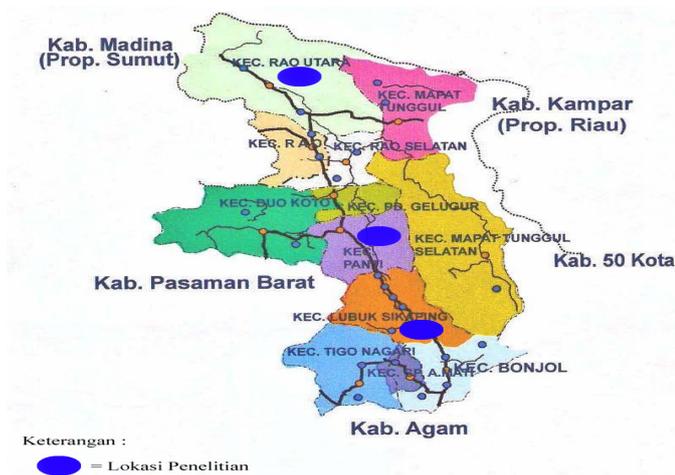


Figure 1. Map of research locations in Pasaman District

Seven morphometrics were measured providing shoulder height (SH), hip height (HH), body length (BL), hip width (HW), chest width (CW), chest depth (CD), and chest circumference (CC). Average values of each body size among buffalo subpopulations were compared by Duncan Multiple Range Test. Age and sex were corrected in determining similarity degree and phylogenetic relationship among subpopulations.

Mahalanobis distance was analysed by Proc. Discrim. of SAS Package (ver. 7.0). Results of calculated quadratic distances were rooted to obtain genetic distance in unquadratic values. Phylogenetic tree was estimated by a simple discriminant function (Herrera *et al.*, 1996), through the approach of a Mahalanobis distance (Nei, 1987) by pooling variant and covariant matrix among variables.

Phylogenetic tree was estimated by Unweighted Pair Group Method with Arithmetic (UPGAMA) using the MEGA program (Kumar *et al.*, 1993). An assumption was given that the rates of evolution within and among subpopulation were similar. Common and mixture values of within and among subpopulations used Canonical analysis (Herrera *et al.*, 1996).

## Results and Discussion

### Pasaman District

Pasaman District is one of central area for production of local swamp buffalo in West Sumatra Province. The location is in the northernmost of this Province for the wide area of around 3,947 km<sup>2</sup> at an altitude of between 50-2240 m asl. The three subdistricts of Lubuk Sikaping, Pantan and North Rao are located in mountain area at the altitudes of 2,340, 1,521 and 1,886 m asl respectively. Cool temperatures of the mountain area are potential in producing good forages. According to the Central Bureau of Statistics of the Pasaman District (2008), Lubuk Sikaping, Pantan and North Rao have area for planting forages successively 2,679 Ha, 4,297 Ha and 2,364 Ha that could accommodate animal respectively 1,181, 979 and 1,134 AU.

### Buffalo Population

Buffalo population in Lubuk Sikaping, North Rao and Pantan were successively around 162, 505 and 140 ST. Population growth was influenced by many factors, such as climate, land availability, management and livestock ownership. Buffalo farmers in North Rao mostly raised their own animals (92%), otherwise farmers as keepers were found in Pantan (63%) and Lubuk Sikaping (58.3%). Ownership status affected the attention of farmers in keeping animals. Farmers in the two latter were very attentive in keeping animals. This might be caused by the status of them as buffalo keepers and also by rearing animals commonly at a small scale (1-3 heads).

### Characteristic of Body Sizes

Morphometric description of three local swamp buffalo subpopulations from the Subdistricts of Lubuk Sikaping, Pantan and North Rao in Pasaman District was presented in Table 1. Data of body sizes were corrected to the same age (4 - 5 years). Average values of each body size of those three buffalo subpopulations observed showed that male buffaloes from Lubuk Sikaping and Pantan had similar body sizes ( $P > 0.05$ ), but both were significantly larger ( $P < 0.05$ ) than male buffalo from North Rao.

Table 1. Morphometric measurement (cm) of local swamp buffalo by sex and location in Pasaman District

Parameter	Female			Male		
	LSi (27)	PTi (35)	NRa (46)	LSi(7)	PTi (7)	NRa(14)
Shoulder H	122.0±2.5 <sup>B</sup>	123.7±2.8 <sup>A</sup>	110.2±4.4 <sup>C</sup>	128.9±2.5 <sup>A</sup>	129.7±4.3 <sup>A</sup>	123.2±2.1 <sup>B</sup>
Hip H.	120.0±2.4 <sup>A</sup>	121.6±2.8 <sup>A</sup>	108.1±4.6 <sup>B</sup>	126.4±2.4 <sup>A</sup>	127.6±4.8 <sup>A</sup>	120.9±1.9 <sup>B</sup>
Body L	113.6±4.1 <sup>A</sup>	113.7±3.6 <sup>A</sup>	113.6±4.0 <sup>A</sup>	116.0±2.9 <sup>A</sup>	116.1±3.1 <sup>A</sup>	114.6±1.7 <sup>B</sup>
Hip W	49.3±4.9 <sup>A</sup>	50.7±5.2 <sup>A</sup>	49.1±4.4 <sup>A</sup>	52.7±5.4 <sup>A</sup>	53.7 ±2.4 <sup>A</sup>	49.6 ±2.1 <sup>B</sup>
Chest W	45.2±4.7 <sup>A</sup>	45.9±4.6 <sup>A</sup>	39.7±3.9 <sup>B</sup>	52.9±3.1 <sup>A</sup>	54.6±4.9 <sup>A</sup>	41.8±1.8 <sup>B</sup>
Chest D	68.5±5.5 <sup>A</sup>	68.9±6.0 <sup>A</sup>	56.1±5.6 <sup>B</sup>	75.6±4.8 <sup>A</sup>	77.6±5.2 <sup>A</sup>	65.2 ±3.5 <sup>B</sup>
Chest G	163.1±3.5 <sup>A</sup>	163.9±5.0 <sup>A</sup>	150.6±7.8 <sup>B</sup>	177.7±6.6 <sup>A</sup>	179.7±6.2 <sup>A</sup>	166.3±3.0 <sup>B</sup>

Description: Different superscripts in the same row was significantly different ( $P < 0.05$ ); LSi: Lubuk Sikaping; PTi: Panti; and NRa: North Rao

Female buffaloes from Lubuk Sikaping and Panti generally also had similar body sizes ( $P > 0.05$ ), but they were larger than female buffalo from North Rao ( $P < 0.05$ ). However, the averages of body length and hip width did not differ among the three subpopulations ( $P > 0.05$ ). Only shoulder height was different among each other, by the order from the larger one was Lubuk Sikaping > Panti > North Rao. These results described that phenotypically, buffaloes from Lubuk Sikaping and Panti had similar body sizes, while buffalo from North Rao had smaller body sizes.

### Mixed values of Phenotype

Based on discriminant analysis results, it could be presumed for the existence of similarity value in a population and for the possibility of how large proportion of the mixed values affecting the similarity by one to other populations (Herera *et al.*, 1996). Buffalo having high value of morphometric similarity in a subpopulation meant that the mixing influences of genes from the outside were reduced. Table 2 presented the highest morphometric similarity was identified in buffalo from North Rao, with the similarity degree of 96.67%, and this buffalo had a relatively small mixture by buffalo from Lubuk Sikaping, in an amount of 3.33%.

Table 2. Similarity and mixture values (%) among three buffalo subpopulations in Pasaman District

Subpopulasi	Similarity and mixture values			Genetic distance		
	L. Sikaping	Panti	North Rao	L. Sikaping	Panti	North Rao
L. Sikaping	67.65	32.35	0.00	*		
Panti	35.71	64.29	0.00	0.48838	*	
North Rao	3.33	0.00	96.67	20.96235	24.90860	*

The degrees of morphometric similarity of the two buffalo subpopulations from Lubuk Sikaping and Panti were nearly similar (67.65% and 64.29%), meaning that the two were genetically interfere at a relatively similar level, but they did not get a genetic mixture of buffalo from North Rao (0%).

### Determination of Genetic Distance and Genetic Tree

The values of matrix of genetic distance among three local swamp buffalo subpopulations (Table 2) showed that buffalo from Lubuk Sikaping and Panti had a closer genetic relationship, with a value of 0.48838. Instead, a longer genetic distance was obtained for buffalo from North Rao to those of buffalo from Lubuk Sikaping (20.96235) and from Panti (24.90860). Genetic tree in Figure 2 further described that buffalo from Lubuk Sikaping and Panti had a closer genetic branch compared to that buffalo from North Rao, of which buffalo in North Rao was at a different genetic branch.

Based on genetic distance and phylogenetic tree obtained, it could be stated that a closer genetic relationship between the two buffalo subpopulations in Lubuk Sikaping and Panti due to the closer locations of both subdistricts, compared to North Rao Subdistrict. This condition offered the possibility for breeders in both locations to mate their buffalo together so intensely that resulted a more genetic similarity in them.

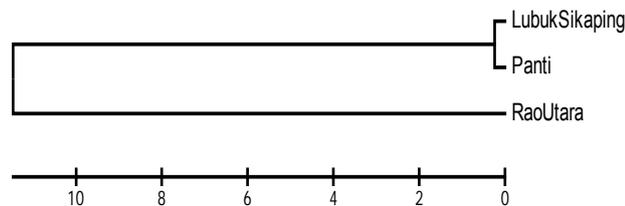


Figure 2. Genetic tree among three buffalo subpopulations in Pasaman District

Efforts for genetic improvement on growth trait of these two buffalo subpopulations could be done through a better breeding program by mating male and female buffaloes as good breeding stocks either from these two locations or from outside Pasaman District. According to Falconer and Mackay (1996) that genetic diversity as expressed in morphology might be caused by mating decision among buffaloes from different locations, beside of selection, isolation, or natural disasters.

For buffalo in North Rao that had a high morphometric similarity value and did not achieve a genetic mixture especially from the two other buffalo subpopulations, in a condition of a conservation action was needed, so North Rao Subdistrict might be considered for in situ conservation. In related to this, Mwacharo *et al.* (2006) stated that in a closed population, from where livestock were reared extensively and unselected, so it might be possible to achieve a significant relationship between geographic difference to genetic distance. Nevertheless, it should be noted also the influence of non-genetic factors in causing morphology differences among local swamp buffalo subpopulations particularly in the three locations in Pasaman District.

## Conclusion

Local swamp buffalo from Lubuk Sikaping and Panti Subdistricts in Pasaman District had similarly body size, but body sizes of both were larger than those of buffalo in North Rao. Local swamp buffalo subpopulations in both locations also had a closer genetic distance compared to that buffalo in North Rao. The nerby locations possibly made an easier way of mixing genetic material between the two buffalo subpopulations through a fairly intensive mating, whereas buffalo in North Rao seemingly had its own genetic purity.

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# Morphometric Comparative Study of Head Linear Surface Measurement of Thin-Tailed, Batur, Wonosobo and Garut Sheep

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## Abstract

Indonesian local sheep have well adapted to the wet tropical climate of Indonesia. The aimed this study to compare the morphometric of the body and head of local sheep. The thin-tailed, Batur, Wonosobo, and Garut ewes (fighting and meat types) at 1.5-2.0 years old were used. Variables of head linear surface measurement observed consisted of akrokranion-prosthion, basion-prosthion, lower jaw length, head height, tuber facial, nasion-rhinion, entorbitale, euryon dan supraorbitale. Data was analyzed using principal component analysis. The principal component chart (clustered diagram) constructed by the components derived from the covariance matrix of variabels measured was created based on the size and shape scores of each linear surface measurement of the body and the head. The results showed that discriminators of head size and shape of Garut sheep (fighting type) was the same, namely head height. The discriminator of the head size was basion-prosthion obtained in thin-tailed and Garut sheep (meat type), that of thin-tailed and Garut sheep (meat type) was basion-prosthion, whereas that of Batur and Wonosobo sheep was akrokranion-prosthion. The discriminator of head shape in thin-tailed sheep, Wonosobo and Garut sheep (meat type) was lower jaw length, whereas that of in Batur sheep was head height. Clusters of head measurement data on breed Garut sheep (fighting and meat types) separated, also on Batur and Wonosobo sheep. Overlapping of head measurement data found on the clusters of thin-tailed, Garut (fighting type), Wonosobo dan Batur sheep.

Keywords: head, morphometric characteristics, principal component analysis

## Introduction

Thin-tailed sheep or Javanese thin-tailed sheep are mostly found in Indonesia (Puslitbangnak 2008). Approximately 80%-85% of this sheep found in West Java and Central Java. Kementerian Pertanian (2011<sup>a</sup>) stated that the Batur sheep was a cross between thin-tailed sheep with Merino sheep with the original distribution in the Batur district that developed since 1974. Phylogeny relationship of Batur sheep was closest to Merino sheep, a bit far with Garut and thin-tailed sheep, most distant with fat-tailed sheep (Prayitno *et al.* 2011). Wonosobo sheep found in several districts in Wonosobo (Setiyawan and Lukiwati 2005). Wonosobo sheep was a crosses between Texel sheep that imported since 1957 with thin-tailed or fat-tailed (Kementerian Pertanian 2011<sup>b</sup>). Garut sheep was developed in 1864, was a cross between thin-tailed sheep with Merino and Cape sheep (possibly Africander sheep of South Africa) (Devendra and McLeroy 1982). Mansjoer *et al.* (2007) reported that Garut sheep was reared as meat and fighting-types.

This study used thin-tailed, Batur, Wonosobo, and Garut sheep (meat and fighting types) at 1.5-2.0 years old. Analysis of body and head morphometrics were based on principal component analysis (PCA) (Gaspersz 1992) then visualized into the group of crowded diagram (Hayashi *et al.* 1982). Different group of crowd builded on the base of scoring in head size and head shape, which derivated from covarian matrix were able to identified morphological phenotypic differences among breeds of sheep studied.

## Materials and Methods

This research was conducted at Jonggol Animal Science Teaching and Research Unit (JASTRU) Faculty of Animal Science, Bogor Agricultural University, Bogor, Batur village Banjarnegara district, Surengede village Wonosobo district and Sindangprabu village Garut district, from from December 2012 to February 2013. The determination of locations was conducted by purposive sampling. The observed sheep were 19 heads of thin-tailed sheep (4 rams and 15 ewes), 26 heads of Batur sheep (7 rams and 19 ewes), 20 heads of Wonosobo sheep (3 rams and 17 ewes), 27 heads of fighting Garut sheep (15 rams and 12 ewes) and 18 heads of meat type Garut sheep (3 rams and 15 ewes). All of the observed sheep were at 1.5-2.0 years old.

The variables of head linear surface measurement observed were *akrokranium-prosthion* ( $X_1$ ), *basion-prosthion* ( $X_2$ ), lower jaw length ( $X_3$ ), head height ( $X_4$ ), *tuber facial* left-right ( $X_5$ ), *nasion-rhinion* ( $X_6$ ), *entorbitale* left-right ( $X_7$ ), *euryon* left-right ( $X_8$ ) dan *supraorbitale* left-right ( $X_9$ ). All variables were measured in head surface of thin-tailed, Batur, Wonosobo, and Garut sheep (fighting and meat types). Data was analyzed using principal component analysis (PCA) with the formula suggested by Gaspersz (1992). Clustered diagram was made based on the head size scores (X axis) and the head shape scores (Y axis).

## Results and Discussion

The equation size and shape of the head surface of thin-tailed, Batur, Wonosobo, fighting-type Garut and meat-type Garut was presented in Table 1. *Basion-prosthion* ( $X_2$ ) was the head size discriminator of thin-tailed sheep with correlation to the head size score of +0912, while lower jaw length ( $X_3$ ) was its head shape discriminator with correlation to the head shape score of +0904. *Akrokranium-prosthion* ( $X_1$ ) was the head size discriminator of Batur sheep with correlation to the head size score of +0969, while head height ( $X_4$ ) was its head shape discriminator with correlation to the head shape score of +0907. *Akrokranium-prosthion* ( $X_1$ ) was the head size discriminator of Wonosobo sheep with correlation to the head size score of +0902, while lower jaw length ( $X_3$ ) was its head shape discriminator with correlation to the head shape score of +0836. Head height ( $X_4$ ) was the head size discriminator of fighting-type Garut sheep with correlation to the head size score of +0905, while head height ( $X_4$ ) was its head shape discriminator with correlation to the head shape score of -0406. *Basion-prosthion* ( $X_2$ ) was the head size discriminator of meat-type Garut sheep with correlation to the head size score of +0928, while lower jaw length ( $X_3$ ) was its head shape discriminator with correlation to the head shape score of +0965. Ozcan *et al.* (2010) stated that the size and shape of the sheep's skull varied depending on the species.

Table 1. Head size and shape equations in the sheep breeds observed

	Thin-tailed sheep
Size equation	$Y_1 = 0.501X_1 + 0.553X_2 + 0.184X_3 + 0.498X_4 + 0.117X_5 + 0.117X_6 + 0.082X_7 + 0.297X_8 + 0.195X_9$
Shape equation	$Y_2 = -0.144X_1 - 0.006X_2 + 0.854X_3 - 0.236X_4 - 0.090X_5 - 0.016X_6 - 0.278X_7 + 0.024X_8 + 0.330X_9$
	Batur sheep
Size equation	$Y_1 = 0.702X_1 + 0.632X_2 + 0.129X_3 + 0.163X_4 + 0.120X_5 + 0.129X_6 + 0.102X_7 + 0.108X_8 + 0.110X_9$
Shape equation	$Y_2 = -0.194X_1 - 0.179X_2 + 0.288X_3 + 0.865X_4 + 0.110X_5 + 0.035X_6 + 0.180X_7 + 0.194X_8 + 0.124X_9$
	Wonosobo sheep
Size equation	$Y_1 = 0.554X_1 + 0.469X_2 + 0.290X_3 + 0.504X_4 + 0.171X_5 + 0.066X_6 + 0.196X_7 + 0.161X_8 + 0.190X_9$
Shape equation	$Y_2 = -0.032X_1 - 0.216X_2 + 0.870X_3 + 0.017X_4 - 0.211X_5 + 0.068X_6 - 0.331X_7 - 0.164X_8 - 0.100X_9$
	Fighting-type Garut sheep
Size equation	$Y_1 = 0.440X_1 + 0.582X_2 + 0.029X_3 + 0.596X_4 + 0.098X_5 + 0.074X_6 + 0.105X_7 + 0.265X_8 + 0.125X_9$
Shape equation	$Y_2 = 0.556X_1 + 0.225X_2 + 0.253X_3 - 0.737X_4 + 0.046X_5 - 0.007X_6 + 0.093X_7 + 0.114X_8 + 0.095X_9$
	Meat-type Garut sheep
Size equation	$Y_1 = 0.507X_1 + 0.580X_2 - 0.003X_3 + 0.535X_4 + 0.055X_5 + 0.047X_6 + 0.129X_7 + 0.257X_8 + 0.180X_9$
Shape equation	$Y_2 = 0.229X_1 - 0.018X_2 + 0.907X_3 - 0.078X_4 + 0.076X_5 + 0.222X_6 - 0.087X_7 - 0.236X_8 - 0.020X_9$

*Akrokranium-prosthion* ( $X_1$ ), *basion-prosthion* ( $X_2$ ), lower jaw length ( $X_3$ ), head height ( $X_4$ ), *tuber facial* left-right ( $X_5$ ), *nasion-rhinion*, ( $X_6$ ), *entorbitale* left-right ( $X_7$ ), *euryon* left-right ( $X_8$ ), *supraorbitale* left-right ( $X_9$ )

Clustered data of sheep breeds observed in Figure 1 showed separation clusters between sheep breeds of fighting-type Garut and meat-type Garut sheep, as well as between Wonosobo and Batur sheep. Separation of the clusters happened because of differences in head shape score, indicating that the head to be inherit. This was accordance with statement of Saparto (2004) that the cranium was inherit to the trait of a breed, so that every breed had a different size of cranium.

Clustered data of sheep breeds observed was found overlapping among thin-tailed, fighting-type Garut and Batur sheep, also between thin-tailed and Wonosobo sheep. It showed similarities between the head of the sheep breeds. The similarity of head morphometric of sheep breeds to be inherit as a result of a common ancestor, ie. thin-tailed sheep. According to Mulliadi (1996) fighting-type Garut sheep was the result of a cross between Merino sheep from Australia, Kaapstad sheep from South Africa and thin-tailed sheep. According to *Surat Keputusan Menteri Pertanian* (2011) Batur sheep was a cross between Merino

and thin-tailed sheep. Wonosobo sheep was the result of a cross between Texel and thin-tailed or fat-tailed sheep.

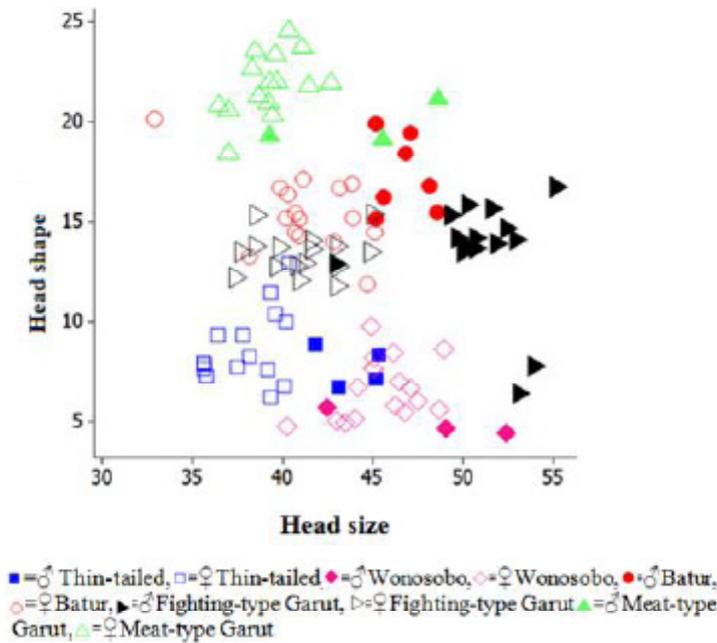


Figure 2. Head clustered diagram of thin-tailed, Batur, Wonosobo, fighting-type Garut and meat-type Garut sheep data

Separation clustered data of head size and shape on fighting-type Garut and meat-type Garut sheep as a result of breeding different directions. Head shape discriminator of fighting-type Garut sheep head was different to meat-type Garut sheep. Head height of fighting-type Garut sheep possibilities associated with gore trait, farmers were indirectly selecting head height as a tool in the ram to defend it self during fighting. Discriminator of head shape was similar to that of head size in fighting-type Garut sheep ie. head height, indicating that this sheep had its own characteristics at head height. Lower jaw length was a head shape discriminator of meat-type Garut sheep that indirectly associated with the ability of sheep to ruminate. Selection wanted specific organs to function in accordance with desired trait.

Overlapping data between fighting-type Garut and thin-tailed sheep because some data from females overlaped each other, but not in males. This indicated that the selection in the direction of fighting-type Garut sheep was clearly visible on head of a male. Differences in the direction of selection in fighting-type Garut and meat-type Garut sheep resulted in the differences in discriminators of head size and shape.

Head size discriminator of Batur and Wonosobo sheep, which indicated that the two breeds of sheep had similar direction (meat-type sheep). Separation clustered data of these two breeds happened due to differences in head shape, each characterized by a different discriminator. Aggressiveness trait of fighting-type Garut was also found in Batur sheep. This was shown by its clustered data which overlapping with fighting-type Garut sheep. Batur sheep closely genetically related to fighting-type Garut sheep (Prayitno *etal.* 2011). In addition to as sheep meat, Batur sheep was kept as pet sheep that often exhibited in the contests.

Lower jaw length was discriminator of thin-tailed, Wonosobo and meat-type Garut sheep. Lower jaw plays a role in the process of taking of food forage and mastication or rumination in sheep. The difference among selection direction between pet sheep (fighting-type Garut or Batur sheep contest) and meat-type sheep indirectly were shown by the difference in quality of feed. Quality of forage in pet sheep was better than that of meat-type sheep. Selection of high quality forage with a low fiber content was given to fighting-type Garut and Batur sheep, so that the function of the lower jaw of these two breeds of sheep was not as good at thin-tailed, Wonosobo and meat-type Garut sheep.

## Conclusion

Head size discriminator of thin-tailed and meat-type Garut sheep was *basion-prosthion*, while that of Batur and Wonosobo sheep was *akrokranion-prosthion*. Head shape discriminator of thin-tailed,

Wonosobo and meat-type Garut sheep was lower jaw length, while that of Batur and fighting-type Garut sheep was head height. Head size and shape discriminators of fighting-type Garut sheep were the same, namely head height. Clustered data of head size and shape separated between fighting-type Garut and meat-type Garut sheep and between Batur and Wonosobo sheep. Clustered data of head size and shape of thin-tailed overlapped with fighting-type Garut, Wonosobo and Batur sheep, while meat-type Garut separated from thin-tailed sheep.

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# Hypoosmotic Test in Rabbit Spermatozoa

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## Abstract

The study was conducted to investigate plasma membran integrity in rabbit spermatozoa, using hypo-osmotic swelling test (HOS test). Six sexually mature rabbit consist of three Lop and three Rex breed used as sperm donor. Semen was collected by using artificial vagina and evaluated macro and microscopically. Semen evaluation included semen volume, pH, consistency, color, mass movement, motility, individual scores, concentration, and viability. Hypo-Osmotic solution was modified by mixing fructose and sodium citrate into three different osmotic pressure: 50, 100, and 150 mOsm/L. Sample of semen (50 µL) in 1 ml HOS solution incubated at 37°C for one hour. Typical tail abnormalities indicative of swelling evaluate every 15 minutes. Data were analyzed statistically using completely randomized design (CRD) with three factors. The result showed that mass movement, motility, and individual scores of Rex's sperm was superior compare than Lop's, but there were no difference in other parameters. The response of plasma membrane integrity showed that Rex's sperm was faster (15 minute) compare to Lop's (30 minute). This fact may relate do the different of plasma membrane composition. In conclusion, to conduct the HOS test in rabbit sperm shoud consider the type of breed.

**Keywords:** lop, membrane integrity, rabbit, rex, semen, sperm

## Introduction

Sperm membrane is fundamental importance in the fertilization process, more attantion has been dedicated to this area of study recently. Two simple test have been available to evaluate membrane integrity; supravital stain and hipoosmotic swelling (HOS) test (Neild *et al.* 2000). When expose to HOS solution functionally active spermatozoa will undergo swelling to establish osmotic equilibrium between the fluid compartmen within the spermatozoon and the extracelluler environment producing the typical swelling of the tail (Neild *et al.* 2000; Amorim *et al.* 2009).

There are references for the use of HOS test in several farm animals such as in equine (Neild *et al.* 2000), Buck sperm (Fonseca *et al.*, 2005), Ram sperm (Nalley and Arifiantini, 2013), and Rabbit sperm (Amorim *et al.*, 2009; Daader and Seleem, 2005)

Since there are plasma membrane composition among breed and individuals, this research conducted to investigate plasma membrane integrity in Lop and Rex breed spermatozoa, using hypo-osmotic swelling test (HOS test).

## Materials and Methods

Six rabbits (3 Rex and 3 Lops) age 1.5-2 years, were used as a sperm source. The rabbit were allocated individually cage and fed a diet providing 100% of their nutrition needs and provide water ad libitum. The semen was collected by using artificial vagina and evaluated macro and microscopically. Semen evaluation including semen volume, pH, consistency, color, mass movement, motility, individual scores, concentration, and viability.

Hypo-Osmotic solution was modified by mixing fructose and sodium citrate into three different osmotic pressure: 50, 100, and 150 mOsm/L. Sample of semen (50 µL) in 1 ml HOS solution incubated at 37°C for one hour. Typical tail abnormalities indicative of swelling evaluate every 15 minutes. Data were analyzed statistically using completely randomized design (CRD) with three factors.

## Results and Discussion

The characteristic of ejaculates collected for analysis are reported at Table 1. Means value of rex was greater than Lop, however in agreement with standard reproductive performance of rabbits.

Table 1. Semen characteristic of Lop and Rex rabbits (means±SD)

Semen variable	Breed	
	Lop	Rex
Volume (ml)	0.47±0.23	0.44±0.24
Color	Creamy white	Creamy white
pH	7.28±0.42	7.37±0.12
Consistency	Watery to thick	Watery to thick
Mass movement	2.33±0.71 <sup>a</sup>	2.67±0.50 <sup>b</sup>
Sperm motility (%)	40±9.35 <sup>a</sup>	61.67±12.58 <sup>b</sup>
Individual score (0-5)	3.67±0.50 <sup>a</sup>	4±0.43 <sup>b</sup>
Sperm viability	47.94±15.02	61.40±18.10
Sperm concentration (10 <sup>6</sup> /ml)	123.89±107.67 <sup>a</sup>	280.56±237.71 <sup>b</sup>
Sperm morphology normal (%)	87.93±2.06	91.27±3.27

Different superscript in the same row differ (p<0.05)

Sperm morphology of rabbit is similar with bull, ram, buck and boar, and the coiling pattern is not differ than those farm animals. In the present study, the highest proportion of reactive sperm differ among solution osmolarity, breed and incubation times. Sperm appeared to suffer coiling (69.77±32.89%) at 50 mOsm/L after 15 minutes incubation in Rex, while in Lop demonstrated at 30 minutes (55.03±9.29%). At 100 mOsm/L Rex and Lop sperm appeared to suffer coiling after 30 and 45 minutes of incubation. When incubated at 150mOsm/L, Lop and Rex demonstrated comparable number of coiling sperm (36.70 and 34.12 %), in different incubation time. Lop at 45 minutes and Rex at 15 minutes incubation (Table 2).

Table 2. Average of total coiling of Lop and Rex spermatozoa exposed on different solution osmolarity after 1 hours incubation (means ± SD)

Solution osmolarity (mOsm/L)	Breed	Incubation time (minute)				
		0	15	30	45	60
50	Lop	22.42±13.73 <sup>b</sup>	32.09±19.05 <sup>b</sup>	<b>55.03±9.29<sup>a</sup></b>	35.02±4.51 <sup>a,b</sup>	32.28±2.43 <sup>b</sup>
	Rex	25.99±7.45 <sup>b</sup>	<b>69.77±32.89<sup>a</sup></b>	59.52±13.44 <sup>a</sup>	48.63±6.40 <sup>a,b</sup>	37.52±7.99 <sup>a,b</sup>
100	Lop	23.36±5.58 <sup>b</sup>	26.33±5.68 <sup>b</sup>	32.82±3.18 <sup>b</sup>	<b>46.29±5.51<sup>a</sup></b>	44.35±4.96 <sup>a</sup>
	Rex	24.88±9.02 <sup>b</sup>	35.15±18.68 <sup>a,b</sup>	<b>60.16±22.13<sup>a</sup></b>	44.18±8.39 <sup>a,b</sup>	39.79±8.47 <sup>a,b</sup>
150	Lop	18.58±9.22	25.82±7.83	27.37±12.17	<b>36.70±21.25</b>	29.33±13.19
	Rex	14.50±14.40	<b>34.12±31.07</b>	25.86±5.42	19.83±2.29	16.47±2.97

Different superscript in the same row differ (p<0.05)

In a hypo-osmotic solution, liquid is transferred into the cell through the plasma membrane of the spermatozoa. Trying to achieve a balance between intra and extracellular space, the functionally intact membrane swells, beginning with the fibers in the sperm tail. As the membranes swell, the spermatozoa curl and invaginate (Jeyendran *et al.* 1984). Osmolarity of the solution should be sufficient to induce the best effect without lysing the sperms. In rabbit sperm, different solutions and times of incubation demonstrated differ of highest proportion of reactive sperm (Table 3).

Table 3. Optimum incubation time of Lop and Rex sperm at different Hypo-osmotic Solution

	Time (minutes)	
	Lop	Rex
50 mOsm/L	30	15
100 mOsm/L	45	30
150 mOsm/L	45	15

When expose in 50 mOsm/L Lop's sperm demonstrated the highest proportion of reactive sperm at 30 minutes, while in 100 mOsm/L and 150 mOsm/L the highest proportion of reactive sperm at 45 minutes. Rex's sperm demonstrated inconsistent result. When expose in 50 mOsm/L and 150 mOsm/L Rex's sperm demonstrated the highest proportion of reactive sperm at 15 minutes, while in 100 mOsm/L, Rex sperm demonstrated the highest proportion of reactive sperm at 30 minutes. This fact explained that not only osmotic solution influence the reaction of coiling sperm, breed of rabbit also influence the result. Amorim *et al.* (2009) suggested for New Zealand breed the suitable for hypo osmotic evaluation was 60 mOsm/L incubated for 30 minutes.

Our research finding explained that during the HOS procedure Lop sperm are subjected to stress conditions and the test selects cells with the most resistant membranes. The lower hypo-osmotic solution, the faster coiling reaction. For Rex's sperm, need to be done using different hypo-osmotic solution composition.

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# Effect of Freezing on Bovine Sperm Morphology

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## Abstract

The evaluation of sperm morphology is an important component of the spermio-gramme and therefore of the andrological evaluation. Cryopreservation processes will reduce sperm motility, acrosome intact, viability as well as sperm morphology. This research aimed to study the effect of cryopreservation to bovine sperm morphology. Six bull consist of two Friesian Holstein (FH), two Simmental and two Limousine belong to Lembang artificial insemination centre were used as a semen source. Semen was collected and processed according to artificial insemination center protocol. Primary sperm morphology evaluate by using bright-field microscopy of carbolfuchsin eosin stained dry-mount semen smears and secondary sperm morphology by phase contrast microscopy of wet-mount semen 'fixed' in isotonic formol-saline. Result demonstrated that Limousine had a lower primary and high secondary sperm abnormality ( $p < 0.05$ ) of raw semen compare to FH and Simmental bulls. Cryopreservation processed increase primary sperm abnormality of Simmental bull and secondary sperm abnormality of FH bull. In total the increasing of sperm abnormal morphology during cryopreservation of three breed were between 1.14 to 1.31%. This research concluded that the cryopreservation processed in Artificial insemination center did not affected on the sperm morphology.

Keywords: frozen semen, morphology, raw semen, sperm abnormality

## Introduction

The evaluation of sperm morphology is an important component of the spermio-gramme and therefore of the andrological evaluation (Al-Makhzoomi *et al.*, 2007). Sperm abnormalites were classified as being primary (head abnormalites) or secondary (tail abnormalities), with the underlying assumptions that primary abnormal (considered to be caused during spermatogenesis) were more serious than secondaries (caused after sperm realese from tubuli seminiferi). Morphologically abnormal sperm can reduce rates of fertilization and embryonic development. It has been generally accepted that bull semen classified as satisfactory should contain at least 80% morphologically normal sperm, with no more than 20% of sperm with an abnormal head. Morphologically abnormal sperm can reduce rates of fertilization and embryonic development (Sarder 2004).

Cryopreservation of semen is routinely used in farm animals, this processes will reduce sperm motility, acrosome intact, viability as well as sperm morphology (O'Connell *et al.* 2002). In Human sperm, O'Connell *et al.* (2002) reported freezing did not significant change in the morphologies of head, midpiece and acrosome in general, except morphologic abnormalities of the tail increased significantly. Apu *et al.* (2012) reported a decrease of total sperm abnormality after freezing in goat semen from from 9.14±0.13 to 13.87±0.38%. Since Indonesia has 15 Artificial insemination centre, this research aims to study the effect of cryopreservation to bovine sperm morphology at one of artificial insemination centre in Indonesia.

## Materials and Methods

Six bull consist of two Friesian Holstein (FH), two Simmental and two Limousine belong to Lembang artificial insemination centre were used as a semen source. Semen was collected and processed according to artificial insemination center protocol. Primary sperm morphology of fresh and after freezing evaluate by using bright-field microscopy of carbolfuchsin eosin stained dry-mount semen smears and secondary sperm morphology by phase contrast microscopy of wet-mount semen 'fixed' in isotonic formol-saline.

Comparison morphology before and after freezing was analyzed by one tailed Students T test using statistical software. Data are presented as means and standart deviation,  $P < 0.05$  was considered significant.

## Results and Discussion

In this research nine type of primary sperm abnormality were found in all bulls ie narrow, tapered, pear shaped, abnormal contour, knobbed acrosome (KA) defect, round head, macrocephalus, microcephalus, and detached headin different number of each bulls. About eight secondary sperm abnormality were detected such as distal midpiece reflect (DMPR) abnormality, bowed midpiece, segmental aplasia (SA) mitochondrial, bent principal piece (bent pp), coiled principal piece (coiled pp), abaxial tail, double tail and teratoid forms.

Raw semen (before freezing) of FH bull demonstated  $7.09 \pm 1.08\%$  sperm abnormalities, and after freezing secondary sperm abnormalities increased  $1.4\%$ , but no deferences was found between raw dan after freezing on the primary sperm abnormalities (Table 1)

Table 1. Sperm abnormalities before and after freezing of FH bull (means  $\pm$ SD)

Sperm abnormalities	Before freezing	After freezing
Primary	$3.53 \pm 0.86$	$3.66 \pm 2.23$
Secondary	$3.43 \pm 1.20^a$	$4.83 \pm 0.59^b$
Total abnormalities	$7.09 \pm 1.08$	$8.36 \pm 0.22$
Total normal sperm	$92.91 \pm 1.87$	$91.64 \pm 2.17$

Note: different lower case letters in superscript in the same raw demonstrate significant differences ( $P < 0.05$ )

Before freezing, Simmental bull demonstated  $6.02 \pm 0.61\%$  of sperm abnormalities, and after freezing  $7.33 \pm 0.19\%$ . Primary sperm abnormalities increased  $0.87\%$ , but no deferences was found between raw dan after freezing on the secondary sperm abnormalities (Table 2). The type of primary sperm abnormatilies was abnormal countour, this can also caused by staining technique.

Table 2. Sperm abnormalities before and after freezing of Simmental bull (means  $\pm$ SD)

Sperm abnormalities	Before freezing	After freezing
Primary	$3.06 \pm 0.62a$	$3.93 \pm 0.74b$
Secondary	$2.96 \pm 0.69$	$3.40 \pm 0.28$
Total abnormalities	$6.02 \pm 0.61$	$7.33 \pm 0.19$
Total normal sperm	$93.98 \pm 0.84$	$92.67 \pm 0.75$

Note: different lower case letters in superscript in the same raw demonstrate significant differences ( $P < 0.05$ ).

No significant diferences found in Limmousine bull before freezing and after freezing with total abnormalities after freezing  $8.43 \pm 1.08\%$  (Table 3).

Table 3. Sperm abnormalities before and after freezing of Limmousine bull (means  $\pm$ SD)

Sperm abnormalities	Before freezing	After freezing
Primary	$2.33 \pm 0.60$	$3.17 \pm 1.02$
Secondary	$4.96 \pm 0.63$	$5.26 \pm 0.96$
Total abnormalities	$7.29 \pm 0.79$	$8.43 \pm 1.08$
Total normal sperm	$92.71 \pm 1.04$	$91.57 \pm 1.24$

Our results demonstrate that overall the sperm morphologies of FH, Simmental and Limousine before and after freezing catagories as an excellent morphologies. Perhaps all bull included to a concept of 'good freezers', as postulated by Watson (1995). This is an extremely important issue, as many artificial insemination centre in Indonesia routinely cryopreserve bull semen. This research prove that aproprite extender and freezing technique at Lembang artificial Insemination center did not effect on sperm abnormalities. Since there are about 15 artificial insemination centre in Indonesia, with different variation of freezing technique

or choice of extender and sperm abnormalities affected the fertility, further study needed to evaluate the sperm morphology before after freezing in all artificial insemination centre.

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# Determination of Soy Extract Concentration in Tris Buffer of Frisian Holstein Chilled Semen

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## Abstract

This aim research was to investigate the best soy extract concentration on the quality of Frisian Holstein (FH) bull's chilled semen. Semen were collected from 5 FH bulls belong to Lembang Artificial Insemination Centre using artificial vagina. Immediately after collection the semen evaluate macro-and microscopically. Semen samples with >75% progressive motility and contained <20% sperm with abnormal morphology were used in the experiment. Semen individually divided into 4 tubes and diluted with 2.5%, 3.75%, 5%, or 6.25% Tris soy extract (TSE), diluted semen than store at 5 °C. Sperm motility and viability were evaluate once a day. The results showed that sperm diluted in 2.5% TSE was the highest sperm motility (30.25±8.45%) at 72 hours of storage (day 3) than in 3.75% (18.00±5.12%), 5% (7.17±2.32%) or 6.25% (4.45±1.02%) TSE. No differences was found on the viability of sperm, between all four TSE concentrations at 0 and 12 hours of storage. At 24 until 72 hour of storage, semen diluted with 3.5% TSE significantly higher than other concentration. This research conclude that sperm motility diluted in 2.5% soy extract better than other concentration, while 3.75% soy extract maintain sperm viability better than other concentration.

Keyword: chilled semen, FH bull, soy extract, tris

## Introduction

Artificial insemination (AI) was the first great biotechnology applied to improve reproduction and genetics of farm animals. The widespread use of AI in cattle can partly be attributed to the availability of suitable diluents. The general requirements for semen diluents are: ionic or non-ionic substances to maintain the osmolarity and to buffer the medium; a source of lipoprotein or high molecular weight material to prevent cold shock, such as egg yolk or milk; glucose or fructose as an energy source; and other additives such as enzymes and antibiotics (Vishwanath and Shannon, 2000).

Soymilk powder or more precisely soy extract (brand Melilea) is full of protein with eight essential amino acids, 40% higher than protein in unprocessed plants. Every 100 g of soy extract contains of 23 g protein, 22 kinds of amino acid, omega 6 and omega 3 fatty acids, and Protein Digestibility Amino Acid Score (PDCAAS). In addition, melilea contains isoflavon, calcium, magnesium, iron, potassium, phosphor, selenium, zinc, and lecithin (Koswara, 2009).

The used of soy extract in tris buffer for buck chilled semen was previously reported by Putra *et al.* (2013). In there research 2.5% of soy extract was the best concentration on the sperm motility and viability of buck chilled semen at 72 hours of storage. Soy extract powder is widely available, and it could be found in most domestic public markets. Information and ingredients of Tris buffer diluents for bull semen are widely available as well. This research aims to develop a Tris-soy extract modified diluents, for Frisian Holstein (FH) bulls chilled semen.

## Materials and Methods

This research were conducted at three places; Lembang Artificial Insemination Breeding Center in Bandung; Reproductive Rehabilitation of Reproduction and Obstetric division, Department of Clinic, Reproduction and Pathology, Faculty of Veterinary Medicine, Bogor Agricultural University.

### Media Preparation

Tris buffer was prepared by mixing 30.28 g Tris-hydroxymethyl-aminomethane, with 17.8 g monohydrate citric acid and 12.5 g D-fructose, dissolved in 1000 mL distilled water according to Arifiantini

*et al.* (2006). Four concentrations of soy extract which were; 2.5% TSE (TSE<sub>2.5</sub>), 3.75% TSE (TSE<sub>3.75</sub>), 5% TSE (TSE<sub>5</sub>), or TSE 6.25% (TSE<sub>6.25</sub>), in Tris buffer (W/V) according to Putra *et al.* (2013). The soy extract was weighed separately based on the treatment and dissolved with Tris buffer to reach 100 mL. Tris and soy extract were homogenized using a stirrer, centrifuged at 2000 rpm for 10 minutes; the supernatant was collected and used as diluents finally added with antibiotic.

### Semen Collection, Evaluation and Processing

Semen from five FH bulls were collected using artificial vagina, based on Lembang Artificial Insemination Center standard protocol; only the first ejaculates of each semen collection were used in the experiment. Following collection, individual semen sample was evaluated macros and microscopically including semen volume, pH, mass activity, progressive motility, individual scoring (velocity), viable sperm, sperm concentration, and sperm morphology. Semen samples with >75% progressive motility and contained <20% sperm with abnormal morphology were used in the experiment.

Semen were processed individually; each ejaculate was equally divided into four tubes and diluted with TSE<sub>2.5</sub>, TSE<sub>3.75</sub>, TSE<sub>5</sub>, or TSE<sub>6.25</sub> to reach the total sperm concentration of  $10 \times 10^6$  mL<sup>-1</sup>. The diluted semen were stored at 5 °C (colling box) and transported to the laboratory at Faculty of Veterinary Medicine, Bogor Agricultural University for further examinations. The progressive motility and viable sperm were evaluated every 12 hours for 3 days observation (72 hours).

### Statistical methods

The data were analyzed using analysis of variance, repeated measurement, the soy extract concentration and time of storage were analyzed separately and to test differences the Turkey's test was used to compare treatment means using the statistical software Minitab 14 version. Data were presented as means±SD.

### Results

Semen diluted with TSE<sub>2.5</sub> showed the highest sperm motility (30.25±8.45%) at 72 hours of storage (day 3) than those diluted with other concentration the sperm motility in TSE<sub>3.75</sub>, TSE<sub>5</sub> or TSE<sub>6.25</sub> were 18.00±5.12, 7.17±2.32, and 4.45±1.02%, respectively (Table 1). The decrease on sperm motility occurred in all diluents and observed after 12 hours after storage.

Table 1. Percentage of sperm motility in different soy extract concentration (means±SD)

Storage time (hours)	TSE <sub>2.5</sub>	TSE <sub>3.75</sub>	TSE <sub>5</sub>	TSE <sub>6.25</sub>
0	75.00 ± 4.05 <sup>aA</sup>	75.00 ± 4.05 <sup>aA</sup>	74.50 ± 4.11 <sup>aA</sup>	74.25 ± 3.60 <sup>aA</sup>
12	71.37 ± 3.89 <sup>abA</sup>	71.87 ± 2.58 <sup>aA</sup>	69.37 ± 3.19 <sup>aA</sup>	65.25 ± 2.28 <sup>bB</sup>
24	65.63 ± 3.90 <sup>bcA</sup>	62.25 ± 3.50 <sup>bA</sup>	53.01 ± 8.19 <sup>bB</sup>	42.87 ± 4.88 <sup>cC</sup>
36	58.61 ± 5.64 <sup>cA</sup>	50.75 ± 6.16 <sup>cB</sup>	33.50 ± 5.96 <sup>cC</sup>	25.25 ± 2.24 <sup>dD</sup>
48	46.75 ± 6.47 <sup>dA</sup>	32.13 ± 5.69 <sup>dB</sup>	15.90 ± 2.85 <sup>dC</sup>	12.75 ± 2.05 <sup>eC</sup>
60	39.13 ± 8.10 <sup>dA</sup>	24.90 ± 5.40 <sup>eB</sup>	11.15 ± 1.64 <sup>deC</sup>	8.225 ± 0.58 <sup>fC</sup>
72	30.25 ± 8.45 <sup>eA</sup>	18.00 ± 5.12 <sup>fB</sup>	7.17 ± 2.32 <sup>eC</sup>	4.45 ± 1.02 <sup>gC</sup>

Note: different lower case letters in superscript in the same column demonstrate significant differences (p<0.01); different capital letters in superscript in the same row demonstrate significant differences (P<0.01); TSE<sub>2.5</sub> = TSE 2.5%; TSE<sub>3.75</sub> = TSE 3.75%; TSE<sub>5</sub> = TSE 5% and TSE<sub>6.25</sub> = TSE 6.25%

The semen diluted with 2.5% modified tris soy extract demonstrated the highest sperm motility percentage than those diluted with other tris soy milk concentrations. This fact maybe related with the osmotic pressure of the diluents. The osmotic pressure of 2.5% TSE diluents was 320 mOsmol/kg, while the 3.75%, 5%, and 6.25% of TSE had higher osmotic pressures which were 357, 364 dan 400 mOsmol/kg respectively. The osmotic pressure of FH bull fresh semen was 250-350 mOsmol/kg. Thus, it is not surprising that 2.5% tris soy milk diluents was better than other tris soy concentration. The osmotic pressure of semen extender is important in preserving sperm survival during storage (Soylu *et al.*, 2007).

In general, the percentage of viable sperm in this study showed a similar decrease with the percentage of sperm motility. The percentage of viable sperm was decreased by 3-5% during the first 12 hours of observation; however, there were no significant differences between all four TSE concentrations. At 24, 36,

48 and 72 hours of storage, semen diluted with 3.5% TSE showed the best viable sperm than those diluted with 2.5%, 5% and 6.25% TSE (Table 2).

Table 2. Percentage of viable sperm in different soy extract concentration (means±SD)

Storage time (hours)	TSE <sub>2.5</sub>	TSE <sub>3.75</sub>	TSE <sub>5</sub>	TSE <sub>6.25</sub>
0	88.72 ± 2.50 <sup>aa</sup>	88.97 ± 2.38 <sup>aa</sup>	89.25 ± 2.54 <sup>aa</sup>	89.22 ± 2.54 <sup>aa</sup>
12	83.96 ± 3.08 <sup>ba</sup>	85.69 ± 1.21 <sup>ba</sup>	85.02 ± 0.90 <sup>ba</sup>	84.01 ± 3.36 <sup>ba</sup>
24	76.29 ± 2.67 <sup>cb</sup>	81.25 ± 1.59 <sup>ca</sup>	79.96 ± 1.52 <sup>ca</sup>	79.29 ± 4.20 <sup>baB</sup>
36	71.04 ± 3.59 <sup>db</sup>	76.58 ± 2.48 <sup>da</sup>	75.66 ± 1.35 <sup>da</sup>	72.52 ± 4.49 <sup>caB</sup>
48	66.74 ± 3.18 <sup>ec</sup>	71.82 ± 1.98 <sup>ea</sup>	71.07 ± 1.32 <sup>eaB</sup>	67.44 ± 4.41 <sup>cdBC</sup>
60	62.70 ± 2.64 <sup>fb</sup>	67.52 ± 1.46 <sup>fa</sup>	66.21 ± 2.16 <sup>faB</sup>	62.42 ± 4.36 <sup>db</sup>
72	57.29 ± 3.84 <sup>sb</sup>	63.01 ± 1.70 <sup>sa</sup>	60.59 ± 3.49 <sup>saB</sup>	56.88 ± 3.74 <sup>eb</sup>

Note: lower case letters in superscript in the same column demonstrate significant differences ( $p < 0.01$ ); different capital letters in superscript in the same row demonstrate significant differences ( $P < 0.01$ ); TSE<sub>2.5</sub> = TSE 2.5%; TSE<sub>3.75</sub> = TSE 3.75%; TSE<sub>5</sub> = TSE 5% and TSE<sub>6.25</sub> = TSE 6.25%

Base on the result the best concentration of soy extract on the motility was 2.5% in the otherhand 3.75% of soy extract was best for sperm viability in the same storage time. In buck semen the highest sperm motility and viability were showed by 2.5% soy extract until 72 hours of storage. Although sperm motility is not directly related to the fertilizing capacity, it is one of the most important factors affecting sperm quality (Oberoi *et al.*, 2014). Base on that statement this research concluded that 2.5% TSE was the best concentration for liquid semen presevation. Future study is needed to add more carbohydrat to improve sperm motility.

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# Identification of Uterin Milk Protein (UTMP) Gene in Bali Cattle by Using Direct Sequencing

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## Abstract

*Serpin peptidase inhibitor, clade A (Alpha 1 antiproteinase, antitrypsin), member of 14 (serpina 14) is well known as uterine milk proteins (UTMP) gene. Uterine serpins are protein secreted by endometrial epithelium during pregnancy. Servina 14 probably serves an immunoregulatory role to prevent rejection of the fetal allograft. The aim of this research was to identify of UTMP nucleotide polymorphisms in Bali cattle using direct sequencing. The total 60 blood samples of Bali Cattle derived from BPTU Bali in Bali siland (20 heads), BPTU Serading in Sumbawa island (20 heads) and Village Breeding Center in Barru District South Sulawesi (20 heads) were used to evaluate their genetic diversity at UTMP gene using direct sequencing. The forward and reverse data sequences were edited using Bioedit program and alignment analysis was carried out using MEGA5 program. Meanwhile haplotype analysis was performed by DnaSP v5 program. The result showed that partial sequences in exon 5 UTMP gene had 16 haplotypes with the highest number of haplotypes were found in VBC Barru district South Sulawesi (8 haplotypes). Moreover, the highest average of haplotype (h) and nucleotide (p) diversity was found in VBC Barru district South Sulawesi were 0.7949 and 0.0016 respectively. In addition, minisatellite insertions were found in exon 5 UTMP gene fragment on Bali cattle which consist of 5'-CCA GTC ATG AAG AAG GCA GAG GTC GTC GTG CCG GCG AAA -3'. Therefore, haplotype and minisatellite variation in exon 5 UTMP gene fragment are potential marker for reproductive traits in Bali cattle.*

*Keywords: Bali cattle, UTMP gene, nucleotide diversity*

## Introduction

The uterine milk proteins (UTMP) gene known as serine proteinase inhibitor and whose function is not widely known (Tekin et al. 2005). Padua and Hansen, (2010) stated that the serpin peptidase inhibitor, clade A (alpha 1 antiproteinase, antitrypsin), member of 14 (serpina 14) is well known as uterine milk protein gene (UTMP), is a protein secreted by the endometrial epithelium during pregnancy. Leslie et al. (1990) found that UTMP was detected in the uterine fluid and secreted by the endometrium of pregnant cows. However, at present, it is not known whether the bovine UTMP gene is expressed in bovine tissues other than the endometrium investigated the distribution of UTMP transcripts in a wide range of fetal and adult bovine tissues (Khatib et al. 2005). UTMP gene expression found predominantly in reproductive tissues and play an important role in reproductive success traits that it was located on chromosome 21 (Khatib et al. 2007), which consists of five exons (Ulbrich et al. 2009). It was further reported, the diversity of UTMP genes significantly affect the future of productive life in Holstein cow due to its important role in supporting the conception during pregnancy (Ing et al. 1989).

An Indonesia native animal genetic resources, Bali cattle has advantages in reproductive trait (Talib et al. 2003). It showed high reproductive capacity (high fertility and calf each year over a long time) (Entwistle and Lindsay 2003), higher reproductive efficiency than the cross-bred (Personal et al. 2015) and high conception rate in different regions (Australia, Malaysia, Bali and Timor) (McCool 1992). Bali cattle in Northern Australia and Sulawesi has a high of 80% to 90% conception rate (Kirby, 1979) and a moderate annual calving rate of from 52% to 67% (Talib et al. 2003). Until now, the information of the genetic characteristics of Bali cattle especially UTMP gene as a potential gene for reproductive traits were still limited. Therefore, the aim of this study is to identify diversity on exon 5 UTMP gene fragment in Bali

cattle by direct sequencing technique. This study is important to design the strategy of Bali cattle breeding program in the future.

## Materials and Methods

A total DNA sampel of 60 blood samples of Bali cattle from BPTU Bali in Bali island (20 heads), BPTU Serading-Sumbawa NTB (20 heads) and Village Breeding Center (VBC) Barru South Sulawesi (20 heads) were extracted using DNA extraction kit (GeneAid). Primer used to amplify the exon 5 UTMP gene fragment based Khatib et al. (2007) was 5'- GGC CCT TCA ACA AGC TGA GA -3' (forward) and 5'- CTA GGG CTC TTG AAC GTT GA -3' (reverse) with length of PCR product 289 bp (Figure 1).

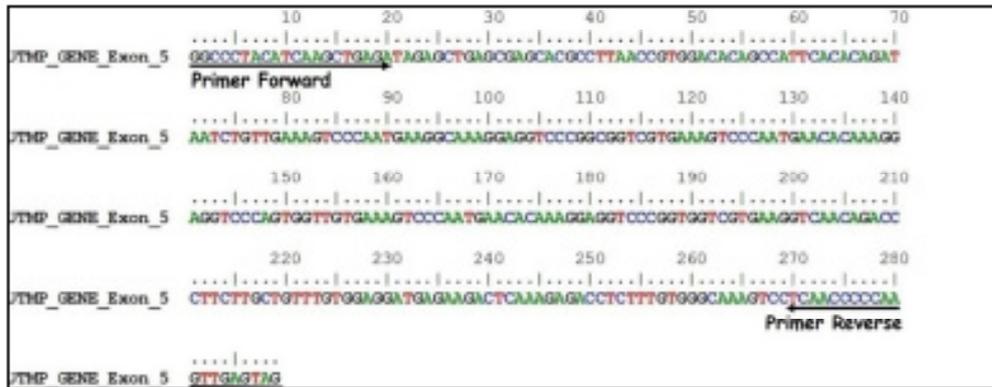


Figure 1. Amplification primer (underline) in the sequence of the exon 5 UTMP gene fragment (GenBank access number L22095).

Amplification of exon 5 UTMP gene fragments used thermocycler AB system machine added by PCR reagent (50 µL) containing 10 µL buffer 10x, 2 µL MgCl<sub>2</sub>, 0.5 µL of dNTP, 0.5 µL of forward primer and 0.5 µL of reverse primer, 0.2 µL of taq polymerase and distilled water 39.3 µL. PCR machine was performed with this following temperature condition: predenaturasi 95 °C for 5 min, 35 cycles (denaturation 95 °C for 10 seconds, annealing 60 °C for 20 seconds, and extension 72 °C for 30 seconds), post extensions 72 °C for 5 minutes, elongation on 72 °C for five minutes in one cycle. PCR products were electrophoresed using 1.5% agarose gel. Furthermore, PCR products were sequenced using the machine sequencer (ABI Prims 3100-Avant Genetic Analyzer) in the 1st Base sequencing company, Selangor, Malaysia. The forward and reverse of exon 5 UTMP gene fragment sequences were edited using Bioedit (Hall, 1999) and MEGA5 programs (Koichiro et al. 2011). Haplotype analysis was carried out by DnaSP v5 program (Librado & Rozas, 2009).

## Results and Discussion

### Amplification of exon 5 UTMP gene

Temperature optimum of exon 5 UTMP gene fragment in Bali cattle was obtained at 60 °C for 20 seconds. Figure 2 showed that the PCR products of exon 5 UTMP gene fragment in Bali cattle (450 bp) were different from the PCR product reference (289bp with Genbank access number L22095). PCR product of Bali cattle was longer than the one of reference because there was an insertion mutations in Bali cattle PCR product. Li and Graur (1991) stated that the insertion is one type of mutation that increases one or more nucleotide in DNA sequence.

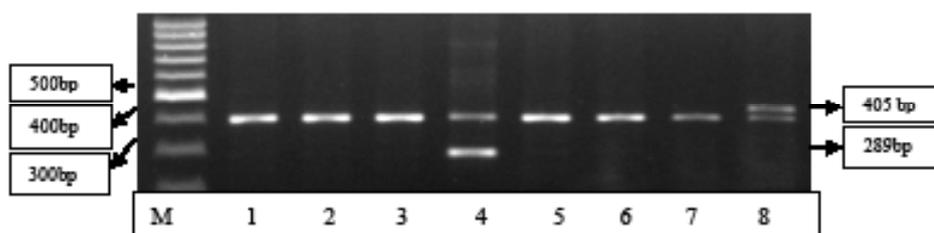


Figure 2. PCR product Electrophoresis of exon 5 UTMP gene in Bali cattle. M is marker of 100 bp, line 1-8 is sample of beef Bali.

### Nucleotide diversity of exon 5 UTMP gene

Haplotype analysis identified 16 haplotypes in the exon 5 UTMP gene fragment from Bali cattle. They were distributed in BPTU Bali cattle in Bali Island (5 haplotypes), BPTU Bali cattle in Serading-Sumbawa NTB (5 haplotypes) and VBC Barru South Sulawesi (8 haplotypes) as presented in Figure 3. A total 16 haplotypes containing around 50 single nucleotide polymorphisms (SNPs) found in three Bali cattle populations may be used as a potential new candidates marker for reproductive traits in cattle Bali (Figure 3). The number of haplotypes (H), haplotype diversity (h) and nucleotide diversity ( $\pi$ ) of Bali cattle are presented in Table 1.

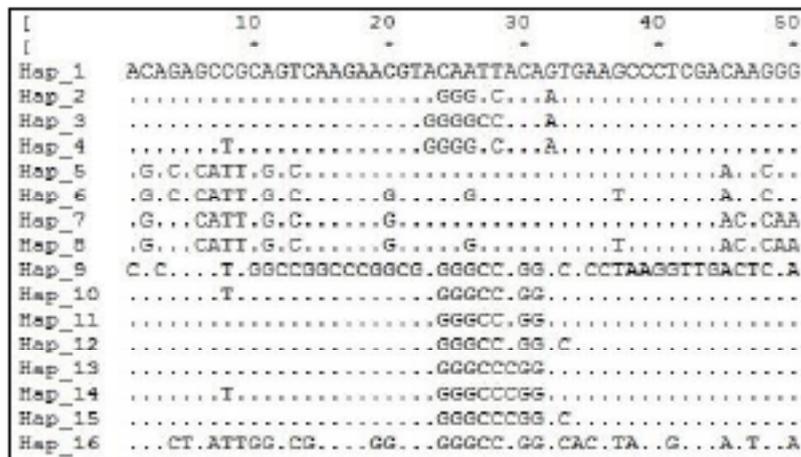


Figure 3. The haplotype diversity of exon 5 UTMP gene fragment in Bali cattle.

Table 1. Exon 5 UTMP gene diversity in three Bali cattle populations

Bali cattle population	n	H	h	p
VBC Barru district South Sulawesi	20	8	0.795	0.016
BPTU in Bali island	20	5	0.585	0.005
BPTU in Sumbawa NTB	20	5	0.708	0.010

n: individual number, H: number of haplotype, h: haplotype diversity,  $\pi$ : nucleotide diversity

The highest number of haplotypes was found in the Bali cattle population from VBC Barru South Sulawesi. Similarly, the highest value of the haplotype (h) and nucleotide ( $\pi$ ) diversities were detected in Bali cattle population in South Sulawesi VBC Barru, they were 0.795 and 0.016, respectively. Haplotype and nucleotide diversity of exon 5 UTMP gene which were found in Bali cattle population is very important. Hall and Bradley (1995) stated that maintaining genetic diversity is the key to long-term survival of the species or breeds. Goodall-Copestake et al. (2012) also reported that the genetic diversity is an important as a reference for the conservation and management programs.

Minisatellite motif insertions in exon 5 UTMP gene fragment with sequence nucleotide motif 5'- CCA GTC ATG AAG AAG GCA GAG GTC GTC GTG CCG GCG AAA -3 'was found from sequencing analysis (Figure 4). Jeffreys et al. (1994) reported that the classification between microsatellites (2-6 bp), minisatellites (up to 100 bp), midisatellites (100-400 bp), and macrosatellites (up to several thousand bp) were based on the number of nucleotide sequences. It is clear that the length of PCR products difference between the this study (405 bp) the one of fragment reference (289 bp) were come from variations of minisatellite in exon 5 UTMP gene. Insertion minisatellite in exon 5 UTMP genes may influence the expression of the produced protein and the possible effect on the gene function. Amino acid sequences generating minisatellite insertion are VPMKAKEVPAVVK. The uniqueness of minisatellite insertion sequences in the exon 5 UTMP gene were only found in Bali cattle population and it was not found in GenBank reference sequences such as reference sequences in UTMPBT201006 Simmental, BT301003 Liomusin and L22095 access numbers. The diversity of minisatellite marker could be used as a genetic marker in the study of population genetics and gene mapping (Jeffreys et al. 1985).

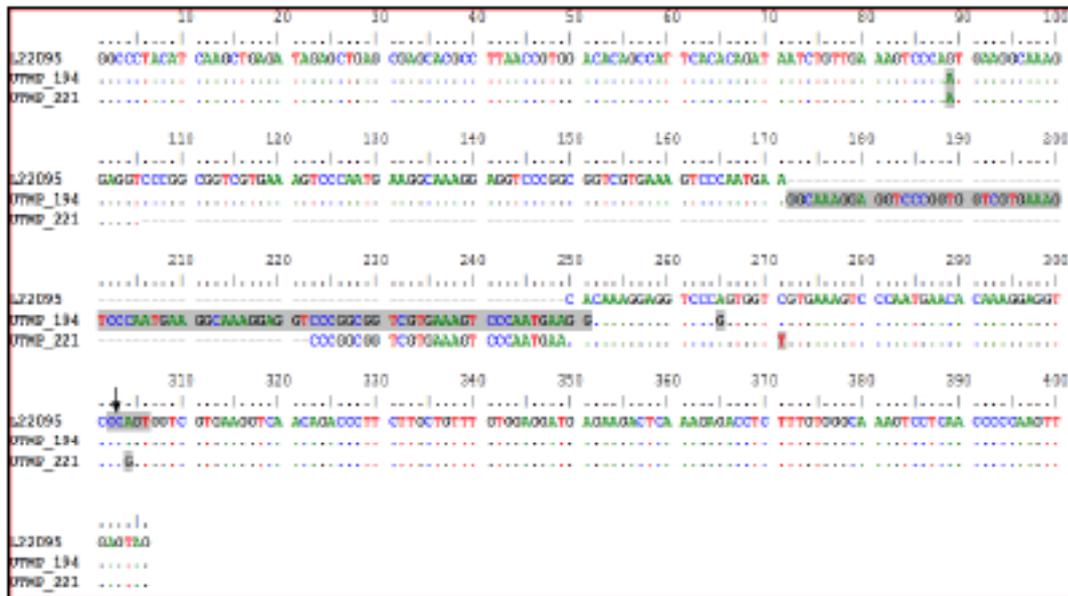


Figure 4. Insertion of minisatellite sequence motif in the exons 5 UTMP gene.

Genetic diversity of exon 5 UTMP gene utilization has been reported by Khatib et al (2007), namely mutation c1.779A>G and c1.296A>G associated with the increase of the lifetime of productive FH cattle. In addition, the GG genotype has lower embryos survival than GA and AA genotypes. Ing et al. (1989) stated that gene expression UTMP was controlled by progesterone synthesis and predominantly found in the endometrium playing a role in maintaining pregnancy. UTMP gene expression also plays as important factor in the productive living age of dairy cattle (Khatib et al. 2009). Genetic diversity particularly haplotype diversity and minisatellite obtained in exon 5 UTMP gene in Bali cattle can be used as a marker for reproduction marker candidate. However, it is needed to investigate the association between haplotype and reproductive trait. Therefore, the haplotype diversity and minisatellite can be used as a reference in the Bali cattle breeding program strategy in the future.

## Conclusion

Analysis of genetic diversity of exon 5 UTMP gene on Bali cattle found 16 haplotypes and highest haplotype and nucleotide diversity were detected in VBC Barru district South Sulawesi. This study also identified minisatellite motif sequences in exon 5 UTMP gene of Bali cattle. Genetic diversity in exon 5 UTMP gene is a potential marker for reproductive traits especially in Bali cattle.

## Acknowledgement

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**THEME F.**  
**ANIMAL PRODUCT TECHNOLOGY AND LOGISTIC**

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# Weight Loss and Mortality of Broilers during Transportation from Different Distances to Slaughterhouse

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## Abstract

Stress during broiler transportation prior to slaughtering results a common phenomenon of weight loss and mortality caused by among other the travel distance. A research in completely randomized designed was carried out to evaluate the weight loss and mortality of live broilers during transportation from farm to slaughterhouse. Broiler transport from each farm location was 3 times replicated. Readily slaughter broilers at 40 days of age with an average weight of 1.5 kg were transported to the same slaughterhouse from 3 different farm locations with different distances of 50 km, 83 km and 153 km for Bogor, Lebak and Pandeglang, accordingly. Weight loss during transportation from Bogor, Lebak and Pandeglang was  $3.33\% \pm 0.06$ ,  $3.73\% \pm 0.76$  and  $4.50\% \pm 1.13$ , respectively. Meanwhile, broiler mortality rate from Bogor, Lebak and Pandeglang was counted to  $0.26\% \pm 0.17$ ,  $0.74\% \pm 0.56$ , and  $0.46\% \pm 0.35$ , accordingly. Weight loss rate showed linear correlation with the travel distance but not to mortality rate. The mortality rate from higher to the lower was found for transportation from Lebak (83 km), Pandeglang (153 km) and Bogor (50 km).

Keywords : broiler, mortality, transportation, weight loss

## Introduction

Transportation of live broiler to slaughter house in term of distance and duration is well known as critical points concerning animal welfare and productivity (Mitchell and Kettewel 1998). Proper transportation of live broilers to slaughterhouse has to be comfortable for broilers to ensure welfare and to reduce weight loss, morbidity and mortality.

Live broilers may suffer from exposure to climatic condition such as high temperature and high humidity, absence of feed and water during journey as well as transporter velocity, vibration and sound. All these factors and their combination disturb broilers performance. The most challenging factors for broiler during transportation are absence of feed and water and exposure to high temperature and humidity which leads to heat stress result in decreasing of body weight, increasing mortality and enhancing morbidity by lowering carcass weight and quality (Mitchell and Kettewel 2009). Hence, weight loss and mortality can be used as indicator of improper transportation activities caused by catching, loading, travelling and unloading procedures (Vecerek *et al.* 2006).

This research aimed to analyze the impact of the existing live broiler transport condition in Indonesia in different travel distance and duration from farms to slaughterhouse on weight loss and mortality.

## Materials and Methods

Readily slaughter broilers in average of 1.5 kg/head were transported from 3 different farms location in Bogor, Lebak and Pandeglang to the same destination at slaughterhouse in Bogor. The distance to the slaughterhouse was measured 50 km, 83 km and 153 km for farm in Bogor, Lebak and Pandeglang, respectively.

Prior to transport, every 15 broilers were placed in a crate measuring 0.94 m x 0.58 m x 0.27 m for length, width and height, accordingly. A total of 144 crates were placed in a truck in a construction of 9-8-2 crates horizontally, vertically and widely. Three thermohygrometers were placed in front, in the middle and in back parts of the truck to measure ambient temperature and humidity at every 60 minutes during journey. Time of start and arrival was recorded to identify journey duration. Final weight and dead broilers were measured.

This research was completely randomized designed with travel distance as factor. This experiment was run in 3 replications (Mattjik and Sumertajaya 2006). Data was analyzed descriptively.

## Results and Discussion

Transportation of live broilers was carried out according to the actual condition in Indonesia by following a same procedure of catching, loading, journey and unloading. Open sided trucks were used to transport broilers along the available road in Indonesia without any treatment and adjustment to microclimatic condition during journey.

A number of 15 broilers in average of 1.5 kg per head were placed in a crate measuring 0.55 m<sup>2</sup> makes a total of 22.5 kg per crate. Broilers were kept densely in a crate with restricted ambulation space, uneven and curvy road resulting to severe shaking which results hard work for broiler leg muscles to stabilize their normal stand. Crate density played important role for broiler, a homeostatic animal, to dissipate heat from the body (Delezie *et al.* 2007) and crate density has to meet the standard of welfare by considering total weight and age of broilers (Legroom 2000). Recommended stocking density for transportation comfort was 34 kg per m<sup>2</sup> or 17 kg per 0.5 m<sup>2</sup>. This crate dense condition triggered low availability of oxygen that effects respiration to release heat from the body and caused hyperthermia (Nidjam *et al.* 2004). Broilers were directly transported to slaughterhouse without resting time along journey. This condition was stressful to broilers and quicker dehydration was then occurred. Most of transported broilers were arrived in exhausted condition caused by dehydration and loss of energy (Warris *et al.* 1993).

### Journey Route, Distance and Duration

Broilers were transported directly to slaughterhouse without any resting period along journey. Journey route, distance and duration were shown in Table 1.

Table 1. Journey route, distance and duration of broilers transportation 3 different farms location

Journey route	Distance (km)	Duration (minutes)
Bogor to slaughterhouse	50	81-90
Lebak to slaughterhouse	83	215-225
Pandeglang to slaughterhouse	153	240-250

As estimated, broilers from Bogor required least time to reach slaughterhouse (81-90 minutes). Although the distance between Lebak and Pandeglang to the same slaughterhouse in Bogor was almost double (70 km different), the travelling time was relatively less different (215-225 vs. 240-250 minutes) because of different routes of transportation (Table 1). The first one travelled through the route of usual road with certain unpleased road condition. Meanwhile, the latter passed through highway which impact to the travelling duration.

Generally, transportation distance revealed a positive correlation with duration. The longer the distance, the longer the duration was. Transport distance and duration become factors that correlate with climatic condition (Table 2) thus weight loss and mortality of broilers (Table 3).

### Temperature and Humidity

High temperature and humidity in Indonesia was accordingly recorded at 24-34 °C and 60%-90% annually (Yani and Purwanto 2006). This observation was done in June of dry season. BMKG (2013) recorded temperature and humidity from May to June in Banten Province (Lebak and Pandeglang) ranged between 23.3-28.5 °C and 61%-94%, meanwhile in Bogor (West Java) the temperature was recorded to 23.0-30.7 °C and humidity was 63%-90%. Temperature and humidity range during transportation was shown in Table 2.

Table 2. Range of temperature and humidity during broilers transportation

Origin	Distance (km)	Temperature (°C)		Humidity (%)	
		Minimum	Maximum	Minimum	Maximum
Bogor	50	24.2	25.2	80	90
Lebak	83	23.9	29.7	70	89
Pandeglang	153	25.2	29.5	75	89

Broilers transport, in tropical circumstance such in Indonesia, usually takes place late in the evening or early in the morning when the temperature is low to reduce heat stress as the trucks are not properly equipped with climatic adjustment. Many authors reported of detrimental effect of transported broiler at high temperature (Warris *et al.* 2005, Viera *et al.* 2011).

Broilers from Bogor area were transported starting from 5 am and arrived in slaughterhouse at 6-6.30 am (60-90 minutes duration). Meanwhile, broilers from Lebak and Pandeglang started to transport at 1 am and arrived at 7-7.30 am in the same slaughterhouse (240-250 minutes duration). These journey times revealed different climatic record. Lower temperature was recorded in journey from Bogor (24.2-25.2°C) compared to Lebak and Pandeglang in which the temperature began to elevate (23.9-29.7°C) by increasing the radiation. However, at those times, the humidity in Bogor area was higher (80%-90%) than in Lebak and Pandeglang because the farm in Bogor was located in a hilly area with lots of water contained in the air (Table 2). Ambient temperature during observation was higher (23.9-29.7°C) than suggested at 20-21°C. This ambient temperature affected the crate temperature during transportation which should be below 23-24°C (Kettlewell 2009). This high temperature, in turn, affected performance of transported broilers (Table 3).

### Weight Loss and Mortality

Broilers were deprived of food, and sometimes water, before catching and crating. During transportation, feeds and water were also withdrawn as feeding and watering was not effectively applicable because broilers held in crates. This usual procedure aims to improve beneficial at slaughter to ultimate meat hygiene by reducing the contents of the gut relieves pressure on the intestines and minimizing leakage of contents if the gut is accidentally perforated during slaughter/dressing procedures (Warriss *et al.* 1999).

Metabolism process still occurs with or absence of feed and water to meet basal requirement in order to maintain homeostasis. Water restriction was particularly likely with high ambient temperatures when large amounts of water lost through panting. Broilers became hungry and dehydrated in such condition. Broilers used glucose reserve from the body when feed was absent and dehydration occurs rapidly without water intake, as water constitutes to 90% of bodyweight (Chen *et al.* 1983). Both factors led to loss of weight and the rate being rather variable and increased positively with increasing journey distance and duration (Warriss *et al.* 1999). Longer transporting caused heat stress and accelerates the metabolism to a point of depletion of muscle glycogen (Owens and Sams 2000). Higher temperature contributes to more weight loss and mortality (Chen *et al.* 1983). Hence, weight loss was a logic consequence of food and water deprivation. Weight loss in this research (Table 3) was even higher than reported by Warris (2000).

Table 3. Weight loss and mortality rate of broilers after transportation

Distance (km)	Weight lost per truck		Mortality per truck	
	Kg	%	kg	%
50	110.50 ± 2.72	3.33 ± 0.06	5.67 ± 3.79	0.26 ± 0.17
83	113.30 ± 28.16	3.73 ± 0.76	14.67 ± 11.50	0.74 ± 0.56
153	118.30 ± 24.01	4.50 ± 1.13	8.67 ± 4.00	0.46 ± 0.35

Transporting broilers during dry season (June) caused higher mortality rate (0.26%-0.74%) although the transportation was carried out during the night and morning (Table 3). In subtropical climate, transported broilers in summer (high temperature) revealed higher mortality rate (0.42%) than in lower temperature in spring (0.39%), in fall (0.23%) and in winter (0.28%) (Viera *et al.* 2011). However, condition of broilers transportation in tropics was not well documented.

The longer the distance and the duration, the more weight loss and mortality was (Table 3). Mortality is triggered and usually happened because of improper handling at catching and transporting. Suggested temperature for broiler in crate during transportation should be below 23-24 °C and ambient temperature of 20-21 °C (Kettlewell 2009). Vecerek *et al.* (2006) reported mortality rate at 0.15%, 0.30% and 0.4% for distance of 50 km, 51-100 km and 101-200 km, respectively. Temperature above 23 °C in this research played role in increasing mortality rate during transportation and even higher than reported by Petracci *et al.* (2010) and Warris *et al.* (2005) at 0.66% as the temperature reached 23-27 °C.

Absence of food and water increased broiler's deep body temperature progressively. The implication of the progressive increase in body temperature was that the broilers were unable to thermoregulate adequately to maintain their temperature constant causing the development of hyperthermia. Liver glycogen, which

provides a ready source of metabolic fuel in the form of glucose, was very rapidly depleted after 50 minutes food withdrawal. Broilers transported 6 hours had only 43% the amount of glycogen in their livers compared with untransported birds (Warriss *et al.* 1993).

Mortality rate was influenced by journey duration, road condition and temperature along journey. Longer journey time revealed different ambient temperature and relative humidity and within the containers in which the birds were held tended to increase. Unpredictably, mortality of broilers from Lebak (83 km) was higher than from Pandeglang (153 km). Crate density triggered the rate of mortality during transportation. Crate density from Lebak was higher than from Pandeglang because of different broilers bodyweight (1.63 kg vs. 1.56 kg for Lebak and Pandeglang, respectively). This finding was in line with the report of Nidjam *et al.* (2005).

## Conclusion

Weight loss and mortality of transported broilers rate increased with increasing travel duration and temperature.

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# Meat Quality of Marica Goat (*Capra hircus*) Meat Fed Different Protein Level

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## Abstract

*The aim of this study was to determine the quality of Marica goat (*Capra hircus*) meat given level of protein. The study was carried out according to completely randomised design (CRD) that consist four treatment and three replication for each treatment. The treatment was protein level (10%, 12.5%, 15%, and 17.5%) fed to the goats. Twelve goat aged 7-10 months were distributed randomly into treatment groups and reared to the research intensively for 2 months. Muscle legs were analyzed to determine the physical properties of the meat including pH value, Water Holding Capacity (WHC), cooking loss, and shear force. These were observed at 0, 3, 6 and 9 days after slaughtering in aging. The results showed that there protein level factor significantly increased ( $P < 0.05$ ) pH value, WHC, and cooking loss, but decreased the shear force value at 17.5% protein level. The aging duration markedly increased pH and shear force value, but declined cooking loss and WHC values. Feed protein level was no effect to physical characteristics of meat except WHC variable. Aging tended to influenced some physical properties of Marica goat meat.*

**Keywords:** aging, feed protein, Marica goat, meat

## Introduction

Goat meat is a rich source of nutrition and is consumed worldwide, especially in the tropics and developing countries, in large quantities (Park, 1988, 1990). One of the potential indigenous and germplasm from South Sulawesi is Marica goat. This goat is a unique genetic resources, superior and have some potential characters to be used as meat producer with various advantages of such a prolific nature (ability to produce twins), high resistance to pressure of climate change, high resistance to parasites and local disease, high ability to survive on land dry in the dry season with minimal grass feed conditions and low quality (Pamungkas, 2009). Once goats have short reproduction cycle and produce quality meat, they are raised as an extra investment without major labor input by the marginal farmers (Sodiq and Sumaryadi, 2002). It was reported by Budisatria et al. (2010) that economic benefits from goats in Indonesia were about 25% higher than those from the sheep flock.

Goat meat has less esteemed than other kinds of meat because of its strong flavor, but on the basis on nutritive and biological value it is not inferior to other kind of meat (Lee et al., 2008). As a meat type, carcass quality and other meat parameters should be addressed to Marica goat. Factors affecting meat quality are pre- and post-slaughter aspects. One of the pre-slaughter is represented by dietary and post-harvest is aging duration. Diets influenced body condition score, flavor and juiciness of cevon (Mapiye et al., 2010). In beef, aging improved some physico-chemical properties with no negative effect on the sensory parameters (Velloto, 2015). This study evaluated the effect of feed level and carcass aging duration on meat quality of Marica goat.

## Materials and Methods

The study was carried out according to completely randomised design (CRD) that consist four treatment and three replication for each treatment. The treatment was protein level (10%, 12.5%, 15%, and 17.5%) fed to the goats. Twelve goat aged 7-10 months were distributed randomly into treatment groups and reared to the research intensively for 2 months and then slaughtered. The carcass were aged at temperature 2-5°C for 0, 3, 6, and 9 days applied to the carcasses of the goats as observation to evaluate the quality of meat. After aging, leg muscle of carcass were taken as sample and measured its pH, Water Holding Capacity (WHC), meat shear force, and cooking loss.

## pH value

Meat sample (10 g) homogenized in distilled water (90 ml) was used to measure the pH value using pH meter (Ockerman, 1985).

## Water-holding Capacity (WHC)

Water-holding capacity (WHC) of meat samples was determined by using press method (Mallikarjunan and Mittal, 1994) with some modifications. Two filter papers were weighed. 1 g of meat sample was placed between them. The sandwich was then placed in a WHC-measuring m/c (Reliance Enterprise, Kolkata, India) and 100 KPa absolute pressure was applied for 1 min. After withdrawing, the meat was discarded and wetted filter papers were weighed. WHC is then calculated as (Mass of filter papers with pressed juice – Mass of dry filter papers)/ Sample mass and expressed as g water/100 g of meat.

## Shear force

The measurement of meat shear force was by using (Creuzot Dumont) shear force. Shear force was expressed in kg/cm<sup>2</sup>. The lower shear force value, meat is the tenderer. Conversely, the higher shear force value of meat, it is harder (Abustam, 1993).

## Cooking loss

Meat sample (20 g) was placed in polyethylene bag and heated in a water bath at internal temperature of 72°C. Cook-out was drained and the cooked mass was cooled and weighed to determine the weight loss (Kondaiah et al., 1985).

## Statistical Analyses

One-way ANOVA was used to compare the treatments effects. Duncan's Multiple Range Test was set to determined significant differences among mean values. The level of significance was  $P < 0.05$ .

## Results and Discussion

### Meat pH

The effect of feed protein level and carcass aging duration on Marica goat meat pH is presented at Table 1. Generally, pH obtained in this study was higher than other reports. Pena et al (2009) claimed that Criollo Cordobes goat had pH 5.72-5.75 and Anglonubians kid goat was 5.71-5.74. However, similar result with this study was indicated by Wattanachant et al. (2008) that nubian x thai goat and saanen x thai goat had pH 6.5-6.6 and Arain et al. (2010) that goat meat had pH 6,28-6,34. The differences pH among studies could be due to different breed studied.

Table 1. Average of leg cut of Marica goat meat pH as affected by dietary protein level at carcass aging duration

Diet Protein Level	Aging duration				Average
	0 day	3 days	6 days	9 days	
10%	6.54	6.30	6.74	6.32	6.46
12.50%	6.14	6.18	6.35	6.52	6.27
15%	6.29	6.42	6.61	6.29	6.39
17.50%	6.53	6.60	6.52	6.25	6.49
Average	6.37	6.37	6.55	6.34	

### Water Holding Capacity

The result of this study on WHC is shown at table 2. Dietary protein level and duration of aging significantly influenced the WHC value ( $P < 0.05$ ). In this study, goat consuming 15% and 17.5% protein in diet increased WHC value. It might be caused by the ability of meat to retain the meat water. This ability corresponded to the amount of protein consumed by goat. Soeparno (2005) stated that the higher meat protein content the higher ability of meat to hold its water. Carcass aged as long as 6 and 9 dayst tended to decline WHC value of meat. It could be resulted in meat protein denaturation. Soeparno (2005) stated that declining WHC was caused by muscle protein denaturation and resulted in meat protein change and free water between protein molecule decreased.

Table 2. Average of leg cut of Marica goat meat WHC as affected by dietary protein level at carcass aging duration

Diet Protein level	Aging duration				Average
	0 day	3 days	6 days	9 days	
10%	32.91	31.26	36.67	36.63	33.91 <sup>a</sup>
12.50%	28.17	32.80	30.99	25.95	29.68 <sup>b</sup>
15%	35.22	41.28	26.26	35.34	35.27 <sup>a</sup>
17.50%	34.06	48.18	29.81	28.86	36.40 <sup>a</sup>
Average	32.59 <sup>c</sup>	38.38 <sup>d</sup>	30.93 <sup>a</sup>	31.69 <sup>b</sup>	

<sup>a, b, c, d</sup> significantly different at  $P < 0.05$

### Shear Force

Shear force is a assessment of hardness or tenderness of meat. The result of the study is presented at table 3. There was no effect by protein level fed to goat ( $P > 0.05$ ), while aging duration tended to increase the shear force value at 6 days ( $P < 0.05$ ). However, the duration 0, 3 and 9 days had almost similar value of meat shear force. Theoretically, aging will extent the change internal enzyme of meat, mainly calpain, to autolyze and degrade meat protein (Goll et al., 2003). This enzyme activity tenderizes meat. This study most probably could not find meat to be tenderer by aging treatment.

Table 3. Average of leg cut of Marica goat meat shear force (kg/cm<sup>2</sup>) as affected by dietary protein level at carcass aging duration

Diet Protein level	Aging duration				Average
	0 day	3 days	6 days	9 days	
10%	0.78	0.59	0.85	0.61	0.70
12.50%	0.78	0.80	0.82	0.72	0.78
15%	0.80	0.78	0.74	0.80	0.77
17.50%	0.60	0.82	0.68	0.84	0.73
Average	0.74 <sup>a</sup>	0.74 <sup>a</sup>	0.79 <sup>b</sup>	0.73 <sup>a</sup>	

<sup>a, b</sup> significantly different at  $P < 0.05$

### Cooking Loss

Cooking loss is an indicator of meat nutrition correlating to meat juiciness and WHC. WHC value will decrease by increasing of meat juiciness (Soeparno, 2005). This study indicated that diet protein did not influence cooking loss ( $P > 0.05$ ), whereas aging tended to decrease the cooking loos ( $P < 0.05$ ) (table 4). Declining cooking loss by aging might be due to correlating to increasing WHC trend (table 3). Generally, however, this value range was lower than Arain et al (2010) report which cooking loss of goat meat more than 30 percent.

Table 4. Average of leg cut of Marica goat meat cooking loss (%) as affected by dietary protein level at carcass aging duration

Diet Protein level	Aging duration				Average
	0 day	3 days	6 days	9 days	
10%	11.00	10.83	8.00	10.10	10.17
12.50%	18.50	22.33	19.50	15.00	19.15
15%	23.67	18.18	13.75	25.00	20.30
17.50%	24.50	22.16	25.25	28.00	24.80
Average	19.41 <sup>a</sup>	18.37 <sup>b</sup>	16.62 <sup>b</sup>	19.71 <sup>c</sup>	

<sup>a, b, c</sup> significantly different at  $P < 0.05$

### Conclusion

Feed protein level was no effect to physical characteristics of meat except WHC variable. Aging tended to influenced some physical properties of Marica goat meat.

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# Skim Milk Powder Substitution with Soymilk Powder Could Improve Physical properties of Beef Surimi-based Sausage

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## Abstract

Surimi technology has been introduced to non-fish meat to produce an intermediate product. One of common non-fish meat applied surimi technology is beef. Further, this product is used as raw material in meat processing such as sausage. The aim of the study was to evaluate the effect of skim milk powder (SM) substitution with soymilk powder (SP) on physical properties of beef surimi-based sausage. The experimental design used was completely randomized design with four treatments including P1 (6% SM: 0% SP), P2 (4% SM: 2% SP), P3 (2% SM:4% SP), and P4 (0% SM:6% SP). The results showed that the substitution of SM with SP significantly decreased pH, Water water Holding holding Capacity capacity (WHC), hardness, and cooking loss of the sausage. pH, hardness value, and cooking loss of sausage markedly decreased ( $P<0.05$ ) when SM was substituted with all level of SP. The WHC value declined ( $P<0.05$ ) at the proportion of SM and SP were 2% and 4% (P3) and 0% and 6% (P4), respectively. Cooking loss value indicated that the higher substitution level of binder, the lower of cooking loss value of sausage ( $P<0.05$ ). In conclusion, the substitution of SM with SP up to 100% of binder improved physical properties of sausage made from surimi-like materials.

**Keywords:** physical properties, sausage, skim, soybean, surimi

## Introduction

Characteristically, sausages are comminuted processed meat products made from red meat, poultry or a combination of these with water, binders and seasoning, and usually stuffed into a casing and may be cured, smoked or cooked (Essien, 2003). Meat used in sausages making are from fresh meat, although it may be made from intermediate products such as surimi-like material. Surimi is a Japanese intermediate product made from ground fish deboned mechanically and washed by using chilling water repeatedly (Lee, 1984). This technology has been applied to non-fish meat such as beef, goat, and duck to produce gelling meat product (Suharyanto *et al.*, 2009; Mega *et al.*, 2010; Ramadhan *et al.*, 2010).

The quality of product is a reflection of raw materials and process status. It is resulted from the assemblage of proper ingredients in the right proportion (Essien, 2003). Binder material is an ingredient playing an important role in sausage. It functions to form emulsion, increase water holding capacity, give structure of meat product (Dzudie *et al.*, 2002), reduce the cost, and increase consumers acceptability of the product (Abdolghafour and Saghir, 2014).

The common non-meat ingredient used in comminuted meat products is milk protein such as skim milk and whey protein. These materials increase moisture, yield and moisture retention of meat patties (Andic *et al.*, 2010), improve texture and sensory properties (Hung and Zayas, 1992), minimize cooking loss, increase water holding capacity and emulsion stability of the products (Serdarodlu and Deniz, 2004). In some cases, milk protein, however, results in a lower emulsifying capacity of the comminuted meat product (Mittal and Usborn, 1985; Zorba *et al.*, 1995) and decreased strength of fracture, hardness, cohesiveness, and chewiness of the meat patties (Andic *et al.*, 2010). Other than the milk protein, soy proteins are also used in processed meat products. Incorporation of soy protein isolate influences a considerable change in physiochemical, microbiological, sensory and textural characteristic of low fat emulsion sausage (Ahmad *et al.*, 2010), and it may be the possible solution to recent consumers demand for low fat and low cholesterol meat product (Yadav *et al.*, 2013). The use of soy flour in meatball increases the yield of product and consumers acceptability, and decreases cost of production (Odiase *et al.*, 2013).

Due to the cost and practical reason, the use of soymilk powder as an ingredient of sausage may be applied to the surimi-like based sausage. This study evaluated the substitution of skim milk powder (SM) with soymilk powder (SP) on physical properties of beef surimi-based sausage.

## Materials and Methods

### Surimi Preparation

The round of beef muscle obtained from traditional market in Bengkulu was separated from fat and connective tissue and then was sized into 3 cm for mincing by using meat mincer. The minced meat was washed three times by using chilling water (5-10°C) which the last washing used chilling 0,5% NaCl solution. The ration of chilling water to minced meat was 3:1. The washed minced meat was dewatered by pressing it in the screen of linen mesh 0,5 mm manually. This product was known as surimi. Surimi was incorporated 5% sucrose and 0.2% sodium polyphosphat. Finally, the beef surimi was freezed in -10°C for 24 hours.

### Sausage Preparation

The frozen beef surimi was thawed at temperature  $\pm 5^{\circ}\text{C}$  for 60 minutes in a refrigerator. The thawed surimi was incorporated with 10% tallow and 10% cube ice for a minute (mixing I). From first mixing, it was blended with 15% cube ice, 15% wheat flour, 10% sago flour, 2% salt (NaCl), 0.2% sodium polyphosphat, 0.5% pepper powder, 0.3% nutmeg powder, 1.5% garlic powder, skim milk powder (SM) and soymilk poder (SP) for 3 minutes (mixing II). SM and SP blended into batter were based on the treatments applied to the sausage. The treatment 1 (T1) was 6% SM : 0% SP, T2 (4% SM : 2% SP), T3 (2% SM : 4% SP), and T4 (0% SM : 6% SP). Each treatments was replicated 3 times. The batter was stuffed into casing with 10 cm length each sausage unit. Sausage was steamed at 65°C for 45 minutes.

### pH and Water Holding Capacity

Sausage pH was measured by using pH-meter (TOA HM-11p). Five g sample in baker glass was diluted with distilled water up to 50 mL and then homogenized by using mixer for a minute. After calibrating to pH 4 and 7, electrode was inserted into diluted sample, the pH indicator rose on screen. Water holding capacity (WHC) was determined by using Hamm method (Soeparno, 2005).

### Hardness of Sausage

Sausages were measured by using Instron UTM-1140 with load cell 2512-204 and the compression was 2830-013 type model. A response of hardness displayed on scale graph. The hardness value was showed by the highest of graphic and expressed in kg/mm.

### Cooking Loss

Samples were weighted before and after cooking. Total weight loss in sausage after cooking was expressed as cooking loss (Dzudie et al., 2002). The percentage of cooking loss was calculated as follow:

$$\text{Percentage cooking loss} = \frac{\text{uncooked sausage weight} - \text{cooked sausage weight}}{\text{uncooked sausage weight}} \times (100\%)$$

### Statistical Analysis

One-way ANOVA was used to compare the treatments effects. Duncan's multiple range test was set to determined significant differences among mean values. The level of significance was  $P < 0.05$ .

## Results and Discussion

All variables indicated a decreasing trend (Table 1). pH value decreased significantly at T2, T3, and T4 from T1 ( $P < 0.05$ ), whereas sausage incorporated SP (T2, T3, T4) had same pH value. It means that the replacement of SM with SP at least 50% could decrease sausage pH. pH value of all treatments showed lower than other study (Ahmad *et al.*, 2010) and it might be due to low pH of beef surimi used (Suharyanto *et al.*, 2010).

The WHC value markedly declined at T3 and T4 from T1 and T2. Substitution more than 50% of SM with SP resulted in decreasing WHC ( $P < 0.05$ ). Honikel (1987) reported that pH has a profound effect on the physical such as WHC, tenderness, and color of meat. In this study, the pH of sausage lowered by replacement of SM to SP and followed by declining of WHC value. The decreasing WHC might be caused by the lower protein content of soymilk than milk (Hasim and Martindah, 2008).

Table 1. Average of pH, WHC, hardness and cooking loss value of beef surimi-based sausage

Variables	Treatments (SM (%) : SP (%) ratio)			
	T1 (6 : 0)	T2 (4 : 2)	T3 (2 : 4)	T4 (0 : 6)
pH	4.65±0.03 <sup>a</sup>	4.56±0.04 <sup>b</sup>	4.52±0.04 <sup>b</sup>	4.51±0.07 <sup>b</sup>
WHC (%)	89.20±0.38 <sup>a</sup>	89.32±0.71 <sup>a</sup>	88.40±0.42 <sup>b</sup>	78.44±0.32 <sup>b</sup>
Hardness (kg/mm)	0.36±0.03 <sup>a</sup>	0.32±0.02 <sup>b</sup>	0.30±0.01 <sup>b</sup>	0.30±0.02 <sup>b</sup>
Cooking loss (%)	14.56±0.23 <sup>a</sup>	12.30±0.21 <sup>b</sup>	6.67±0.13 <sup>c</sup>	6.52±0.19 <sup>c</sup>

<sup>a-c</sup> different superscript within a row are significantly different (P<0.05)

The response of hardness value had the same pattern to pH value. It decreased at T2, T3, and T4 from T1 (P<0.05). The substitution of SM with SP at least 50% resulted in the hardness of sausage lowered. It indicated that the product with SM substituted by SP was more tender. The decreasing hardness value might be correlating to the lowered WHC value of sausage. This study result is contrary to other finding. The batter of comminuted product made from beef surimi had lower shear strain and emulsion stability. It might be due to low sarcoplasmic protein content in surimi (Farouk *et al.*, 2002). However, Farouk *et al.* (2002) revealed that the cooked batter shear stress was not affected by sarcoplasmic protein, while this study indicated decreasing shear force of cooked sausage.

Cooking loss of sausage significantly decreased from T1 to T2 and more decreased at T3 and T4 (P<0.05). The lower of cooking loss the higher of cooked yield. This finding did not support Farouk *et al.* (2002) that claimed comminuted product-based surimi had low cooked yield. However, other study revealed that addition of soymilk powder increased moisture, yield and moisture retention of patties and did not cause any negative effects on the sensory properties of the meat patties (Andic *et al.*, 2010).

## Conclusion

Soy milk powder could improve some physical properties of sausage made from beef surimi. However, advance study is needed to evaluate differences between surimi-based and fresh meat-based sausage properties. It is also important to confirm some different result of the similar study.

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# Effects of Local Flour Types on Physical Properties and Acceptability of Beef Sausage

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## Abstract

*This research aimed to study the effect of local flour types on the physical properties (cooking loss, tenderness, water holding capacity) and the acceptability (taste, flavor, and texture) of beef sausage which is equal to the control. In this study, the observed types of local resources consisting of tapioca, ganyong flour, palm flour and sweet potato flour on the physical properties and acceptability of beef sausage. This research was carried out experimentally using a completely randomized design with four treatments (flours) of 10% of the weight of the meat, i.e. tapioca as the control (P0), ganyong flour (P1), palm flour (P2) and sweet potato flour (P3) and each treatment was replicated five times for each of the measured variables. To determine the effect of treatment carried out analysis of variance and to know the difference between treatments used Tukey test. The results showed cooking loss and tenderness were not different from controls. Water holding capacity was different only for palm flour were different from control. Acceptability (taste, flavor and texture) was not different from controls for all types of flour used.*

**Keywords:** *acceptability, physical properties, types of flour*

## Introduction

According to Aberle, et al. (2001) the chemical composition of meat is mainly made up of 65-80% water, 16-22% protein, 1.5-13% lipid, 1.5% non-protein nitrogenous substances and 1.0% carbohydrates and non nitrogenous substances. Variations of the chemical composition of meat can be caused by differences in growth, feed, breed, muscle location and storage method. Due to its high protein and lipid content, it is very important to prolong the shelf life by meat processing technology.

One of processed meat which is very popular in society is sausage. The sausage is a food product obtained from a mixture of minced meat with flour or starch with or without the addition of seasonings and other allowable food additives and put into sausage casings (Standar Nasional Indonesia, 1995)

According to Wilson (1981), the stages of manufacture of sausage is as follows: meat mincing, blending, filling to casing, tying, cooking, cooling, cutting and packaging. In the mixing of materials typically used emulsifying machine is a combination of milling and enumeration system. At this stage add ice cubes, salt, binders, fillers and other additives that can be distributed evenly. Cooking sausage dough into sausage casings using a special tool, in order to establish and maintain the stability of sausages and reduce the formation of air pockets that will affect the quality of the sausages (Person and Tauber, 1984). Cooking aims to bring together components of the dough, stabilizing color and disable microbes (Lawrie, 2003).

Manufacturing of sausages requires binder and filler. Binder is a material that capable to bind water and emulsify fat and high in protein, while the filler is a material containing high carbohydrate. Filler is useful for improving the stability of the emulsion, increasing the water holding capacity and flavor and reducing cooking loss during cooking (Pearson and Tauber, 1984). Some of local flours having the potential to be used as filler is ganyong flour, palm flour and sweet potato flour. This research aimed to study the effect of local flour types on the physical properties (cooking loss, tenderness, water holding capacity) and the acceptability (taste, flavor, and texture) of beef sausage which is equal to the control.

## Materials and Methods

The materials used in this research was beef, tapioca, ganyong flour, palm flour, sweet potato flour, ice, salt, garlic, skim milk, nutmeg, pepper and casing.

### Experimental procedure

1. Meat which has been in the deboning weighing 5 kg and then cut into small pieces and then divided

250 g for each treatment is then milled using a food processor for 1 minute.

2. Add the tapioca flour (P0), ganyong flour (P1), palm flour (P2), sweet potato flour (P3), ice cubes, salt, garlic, margarine, skim milk, nutmeg and pepper and then milled again in a food processor until well blended.
3. Then the dough is inserted into the casing with a stuffer, after the sausage tied by a rope to a distance of 6 cm binder.
4. The sausages were weighed to calculate the weight before cooking
5. The sausage is cooked in a temperature of 60 - 75°C for 45 minutes. The sausages were weighed to obtain the weight after cooking. Subsequently, cooking loss was calculated by the following formula:

Cooking loss= (weight before cooking - weight after cooking) / weight after cooking

6. Tenderness measured by the following method: 2 cm sliced sausage was put under the penetrometer needle in such a way the needle touch the sample surface. The penetrometer was then pressed for 10 seconds. This procedure was performed 10 times in the different places of the sample and its value displayed in the monitor. The tenderness was then calculated by:

Tenderness (m/10 seconds/g)= measurement average/10 seconds

7. Water holding capacity was tested by press method according to Hamm method (modification from Zamora G.M, et al 2015 and Suliman A.M. E,et al 2014), namely by charging 0.3 grams of meat samples on filter paper between two plates with a weight of 35 kg. After 15 minutes, the area of meat samples and the wet were marked and measured by planimeter. Wet areas is the area of water absorption on the filter paper after clamped for 5 minutes subtracted by the closed area of meat samples. Water holding capacity is calculated by the following formula:

$mgH_2O = [(Wet\ area\ (cm^2) / 0.0948] - 8.0$

$\% \text{ free moisture} = (mgH_2O / 300\ mg) \times 100\%$

Water holding capacity= total moisture content - free moisture content

8. Acceptability test (including taste, flavour, and texture) used 5 hedonic scale for all those treatments with 20 people semi trained panelis. The hedonic scale used was: dislike = 1, neutral = 2, rather like = 3, like = 4 and most like = 5 (Soekarto, 1993). The design used in this research was complete randomized design with four treatments, tapioca as control (P0), ganyong flour (P1), palm flour (P2) and sweet potato flour (P3) and each treatment was replicated five times. The data obtained from the physical properties and acceptability tests were statistically analyzed by analysis of variance and to investigate the difference between treatments, the Tukey test was used (Steel and Torie, 1984).

## Results and Discussion

The results showed that cooking loss ranged 1.58 - 2.64% and tenderness ranged 189.10 - 218.12 mm / g / 10 sec and water holding capacity ranged 43.27 - 53.81% (Table 1). Results of analysis of variance showed that cooking loss and tenderness provided no significant effect. This was due to the flour used have similar composition and physical. Tapioca flour contains carbohydrates 88.88%, water 10%, protein% 0:51, 0:31% fat, carbohydrates 85.20 ganyong flour, 14% water, 0.7% protein, 0.2% fat, carbohydrates 89.31 palm flour, water 10%, proteins 0:51% , 0:07% fat, and sweet potato flour contains carbohydrates 86.95, water 7.80%, 2.16% protein, 0.83% fat ( Departemen Kesehatan RI, 1981 dan Laboratorium Pascapanen Balai Penelitian Pertanian Bogor,2003)

Table 1. Value of physical quality sausages

Treatment	Cooking loss (%)	Tenderness (mm/g/10 seconds)	Water holding capacity
Tapioca flour (P0)	1.58 a	218.12 a	51.73 a
Ganyong flour (P1)	2.64 a	190.22 a	53.81 a
Palm flour (P2)	2.70 a	211.83 a	43.27 b
Sweet potato flour (P3)	2.24 a	189.10 a	50.61 a

Description: The same letter are not significantly different column direction

In addition to the similar composition and physical properties of the all flours, the amount of flour added to the dough in all treatments have the same percentage namely 10% of the weight of the meat so that the protein content of all treatments were similar. The amount of protein in the dough affect cooking loss which is in line with Teye and Teye (2011), i.e. addition of flour will yield a stable matrix resulting in same cooking loss.

Cooking loss of all treatment were in the general range of cooking loss i.e. 1.5 - 54.5 (Suparno, 2005). Cooking loss is closely related to the water holding capacity of dough. This is in line with the opinion Teye, M and Teye, GA (2011), i.e. the amount of cooking loss is influenced by the amount of water emerging from the meat, protein degradation and the ability of meat to bind water.

Moisture content contributes in cooking loss. As the number of hydrogen bonds in the sausage is higher the less nutrients emerging during the cooking process. Cooking loss is an indicator of the nutritional value of meat associated with the moisture content of meat, which is the amount of water bound in and between muscles. Other factors affecting loss are the sausage cooking starch contained in the carbohydrates. When starch contact with water, the starch granules will absorb water and swell, but the amount of water absorbed and the swelling is limited, only reaching levels of 30% (Winarno, 2004).

The high content of starch added will affect the stability of the emulsion so that all the sausages produced have the same tenderness. Tenderness was affected by moisture, fat and protein therefore increased water holding capacity caused increased tenderness (Ockermen, 1983).

Water holding capacity of the sausage with palm flour significantly different from other types of flour. This resulted in highest cooking loss of the sausage with palm flour. The protein in the dough leave along with the water. Violeta (2012) stated that as the protein content reduces the water holding capacity decreases.

Table 2. Value acceptability sausages

Treatment	Taste	Flavor	Texture
Tapioca flour (P0)	4.00	3.95	3.98
Ganyong flour (P1)	3.36	3.45	3.47
Palm flour (P2)	3.41	3.52	3.37
Sweet potato flour (P3)	3.55	3.67	3.42

Taste is one of factors that greatly affect the consumer acceptance of processed food products. The average score for the criteria of taste ranging from 3.36 to 4.00, which means between rather like to like. Flavor sausages produced in general from all formulations are relatively the same. This is due to the starch do not have taste ( Fardiaz, et al.1992). It is also contributed by the percentage of spice used in all treatments were the same. Seasoning contribute in the formation of sausages flavor and is enhanced by cooking (Kramlich,et al. 1973).

The average score for the flavor ranging from 3.45 to 3.95, which means between rather like to like. In general, the flavor of all the sausages were accepted by the panelists. Each of tapioca, ganyong flour, palm flour and sweet potato flour has a specific flavor but still acceptable to the panelists. All flours used can maintain the protein, resulting in a preferred flavor (Wilson,1981). Flavor of processed meat products are also affected by the age of cattle, feed type, species, sex, fat, breed, storage conditions after cutting meat, the type of cooking, as well as the cooking time and temperature (Lawrie, 2003). Cooking process plays an important role in this characteristics because during the cooking process of fat in the sausage, volatile components contributing to the flavor emerges. This is in line with the opinion Soeparno (2005) that the presence of cooking will produce volatile compounds providing a unique flavor of the processed meat.

The average value for a texture ranging from 3.37 to 3.98, which means between rather like to like. In general, the texture of the sausages were still accepted by the panelists. Texture and tenderness of the food is often determined by the moisture content and fat, as well as the type and structure of the existing carbohydrate and proteins (Violeta, 2012).

All flours used in this study have a good characteristic for sausage's filler. Their starch content were high i.e. 73-80% amilose and 17-24% amilopektin, with gelatinized temperature ranging from 53 to 70°C (Laboratorium Pascapanen Balai Penelitian Pertanian Bogor, 2003 dan Winarno, 2004). Therefore, the physical quality of the sausages made from all flours were similar.

## Conclusion

Cooking loss and tenderness were not different from controls. Water holding capacity was different only for palm flour were different from control. Acceptability (taste, flavor and texture) was not different from controls for all types of flour used.

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# Characteristic of Lactic Acid Bacteria Isolated from *Danke* from Sinjai, South Sulawesi

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## Abstract

“Dangke” is an Indonesian traditional and well known food to the community of South Sulawesi, which is made from buffalo and bovine milk. This product was made using boiled milk added with papain enzyme as coagulated derived from fresh juice papaya. An experiment was conducted to isolate, identify, and then characterized lactic acid bacteria from “Dangke”. A factorial completely randomized design was used in this experiment with different incubation temperature (10°C, 37°C, 45°C) and different isolate Lactic Acid Bacteria (LAB) were the factors of this experiments and parameter that measured was LAB population with three replications. Data was analyzed using analysis of variance (ANOVA) and Tukey HSD. API 50 CH kit was used for LAB identification. The result showed that LAB isolated from Dangke were *Pediococcus pentosaceus* MSS 1 and *Pediococcus pentosaceus* MSS 2. These species of LAB had characteristic of gram positive, tetrad coccus, homofermentative, catalase test negative, and mesophiles LAB that grew optimum at incubation temperature of 37°C and still survived at 45°C. Different incubation temperature had influenced to population of *Pediococcus pentosaceus* MSS 1 and *Pediococcus pentosaceus* MSS 2 on 24 hours growth ( $P < 0.05$ ).

Keywords: Dangke, lactic acid bacteria, *Pediococcus pentosaceus* MSS 1, *Pediococcus pentosaceus* MSS 2

## Introduction

Consumption of animal protein has been increase each year. Department of Agriculture reported that animal product consumption such as meat, egg, and milk increased from 2008 up to 2011. Milk as one of animal protein consumption increased around 14.26/kg/capital/year (Deptan, 2012). Based on SNI 3141-2011, milk is defined as a liquid produced by clean and health udder of mammals which obtained from good milking process and received no treatment except cooling (BSN, 2011).

Milk contained complete and high nutrient. Fresh milk contained 87.5 % of moisture, 5 % of lactose, 3.5 % of protein, and 3-4 % of fat (Widodo, 2002). This highly nutrient content in milk is good medium for bacteria growth, both pathogenic bacteria and spoilage bacteria (Rahayu and Nurwitri, 2012). Bacteria contamination able to grow quickly and caused rotten milk and not feasible to consume. One treatment to reduce the contamination of milk is through processing milk into some product like dangke.

Dangke is an Indonesian traditional food from Sinjai, South Sulawesi made from buffalo and bovine milk. This product has a high nutrient and preferred by consumers. Dangke was processed by boiling the fresh milk then added fresh fruit juice of papaya until forming a curd (Surono and Hardjo 1984). Papain enzyme was a proteolytic enzyme from fresh juice papaya (*Cacica papaya* L) (Yuniwati *et al.*, 2008). Arni (1993) reported that dangke was included in cheese non-ripening dan had high moisture content (63.83%). Hatta *et al.* (2013) reported that dangke made from bovine milk which contained 55 % of moisture, 2.1 % of ash, 14.8 % of fat, and 23.8 % of protein.

Lactic acid bacteria (LAB) played a role as basic material (starter) to enhance shelf life of food, meat, milk, and vegetables through fermentation process (Fardiaz 1989). Various studies reported that LAB of dangke had function as good bacteria for health. LAB of dangke which had succeeded isolated were *Lactobacillus plantarum* DU15, *Enterococci faecium* DU55, and *Leuconostoc mesentroides* DU02 (Razak *et al.*, 2009). Moreover, LAB identification from dangke had been in few numbers, therefore it still needed to identify LAB from dangke. This product had to maintain and conserve due to its contribution in food, especially by LAB identification.

## Objective

The objective of this research was to analyze the physical characteristic of dangke, to isolate and identify LAB of dangke from Sinjai, South Sulawesi.

## Materials And Method

The research was conducted at Integrated Laboratory, Departement of Animal Production and Technology, Faculty of Animal Science, Bogor Agricultural University.

### Materials

Physical characteristic of Dangke was consisted of pH value and total titratable acid (TAT) that used 0,1 N of NaOH and phenolptaline indicator. Growth medium for bacteria was deMan Rogosa Sharpe Broth (MRS Broth), deMan Rogosa Sharpe Agar (MRS Agar), Yeast Extract (YE), and Bacteriological Agar (BA). Gram staining materials consisted of sterile aquadest, sterile aquabidest, immersion oil, alcohol, NaCl, Buffer Peptone Water (BPW), violet crystal, iodine, 95 % of alcohol and safranin. Material for LAB identification was using CHL medium.

### Equipment

Equipments that used in this research, were pH meter, autoclave, incubator, laminar air flow, refrigerator, microscope, vortex, ose needle, micropipet, reaction tube, erlenmeyer, plate, object glass, drop pipet, bunsen, beaker glass, tip, reaction tube rack and other glass, while LAB was identified by using API 50 CHL kit.

### Procedure

#### *Preparation sample*

Sample of dangke was collected from Sinjai, South Sulawesi which made from bovine milk and processed enzymatically use papain from papain juice.

#### *Physical Characteristic (AOAC 2005)*

**pH value.** Five g of dangke sample was weighed and homogenized in 5 mL aquadest then pH sample was measured using electrical pH meter Schott which had been calibrated using pH 7 of standard solution, then calibrated using pH 4 of standard solution. The electrode tip was placed in sample solution until hear a sound. The pH value was noted and the measurements in 3 equal replicates.

**Total Titratable Acid.** Ten g of sample was weighed and added with 50 mL aquadest then homogenized. Sample was added by 2-3 drops of phenopthaline indicator then titratable with NaOH (0.1 N) until pink color showed up. The used of NaOH volume and TAT measurement in 3 equal replicates. Total acid was calculated with the following formula:

$$\text{Total Titratable Acid} = \{[\text{Vol. of NaOH used} \times \text{N NaOH} \times 90.08 \text{ (MW of lactic acid)}] / \text{Sample weight}\} \times 100\%$$

#### *Microbiology Characteristic*

##### *Isolation of Lactic Acid Bacteria (LAB) (Sujaya et al. 2008; Ogunbanwo et al. 2003)*

One ose of Dangke sample was streaked onto solid MRS Agar medium. The plate was incubated at 37 °C for 24 hours. Isolation was continued until pure culture was obtained. This isolate was used for working culture to confirm the bacteria characteristic.

##### *Identification of LAB (Lay 1994; Hadioetomo 1990)*

**Catalase Test.** One ose of isolate from plate was lubricated onto object glass sterilized by alcohol, then dropped with 3 % of H<sub>2</sub>O<sub>2</sub> solution. The preparete was observe, gas bubbles indicated that the bacteria had positive catalase, while no gas bubbles indicated that the bacteria had negative catalase. Each test was repeated 3 times.

**Morphological characteristic.** A half mL of aquadest was dropped onto object glass, then one ose of LAB isolate was lubricated on it. Next, the object glass was covered with cover glass. The preparete glass was dropped with imersion oil then seen under microscope at 100X of magnification.

**Gram Staining.** Bacteria sample from homogen colony (pure culture) was lubricated onto sterile object glass then made thin preparation and heat fixed. The bacteria was dropped with violet crystal solution for 2 minutes then washed with aquadest. Next, the bacteria was dropped with iodium and air dried for 2 minutes, washed with aquadest and dried. The preparete was washed with 95 % of alcohol for 30 seconds,

then immediately washed with aquadest and dried. Furthermore the preparate was dropped with safranin for 30 seconds, washed with aquadest and dried. The preparate glass was dropped with imersion oil then seen under microscope at 100X of magnification to obtained shape of cell and color of cell wall after staining. Gram-positivebacteriawould showed purple or dark blue color, while the Gram-negative bacteria would showed red color of safranin inside the cell wall.

#### ***LAB Identification (Annuk et al. 2003)***

The identified LAB based on morphological, culture characteristic, catalase test, and Gram staining the further identified biochemically by carbohydrate fermentation using analysis of API 50 CHL kit based on the ability to ferment the 49 types of carbohydrate to determine the species of LAB. The choosen isolate was subcultured in MRS Broth medium at optimum temperature for 24 hours. The identificatin procedure was performed based on manual standard book of API 50 CH kit. The observation data was analysed using APIweb software (bioMérieux).

#### **Parameter**

##### ***Preparation of Standar Curve (Kusmiati and Malik 2002)***

One mL of bacteria culture (24 hours) was homogenized with 9 mL of MRS Broth (1:9 comparisons). The homogenate was made as first serial dilution. The the second dilution was made by inoculated 5 mL from first dilution in 5 mL of MRS Broth (1:1 comparison) sequential started from  $\frac{1}{2}$ ,  $\frac{1}{4}$ ,  $\frac{1}{8}$ ,  $\frac{1}{16}$ , and  $\frac{1}{32}$  of dilution. Next, from each dilution was performed terraced ilution using BPW with 1:9 comparisons until  $10^9$  for  $\frac{1}{2}$ ,  $\frac{1}{4}$  and  $\frac{1}{8}$  and also until  $10^8$  for  $\frac{1}{16}$  and  $\frac{1}{32}$  of dilution. Further, the last three dilution was obtained for pour plate technique to determine the population of LAB.

One mL of culture was picked up and poured onto plate then MRS Agar was poured and homogenized. The plate was incubated for 24 hours at 37°C then population of LAB was counted. The optical density at  $\frac{1}{2}$ ,  $\frac{1}{4}$ ,  $\frac{1}{8}$ ,  $\frac{1}{16}$ , and  $\frac{1}{32}$  of dilution started from highest dilution was measured using UV-VIS spectrophotometer at  $\lambda_{600\text{ nm}}$ . Further, the equation of standard curve was obtained as followed:

$$y = ax + b$$

##### Note:

- y : bacteria population
- a and b : value of variables
- x : absorbance

##### ***Growth of LAB at 10 °C, 37 °C, and 45 °C***

One mL of working culture (24 hours) inoculated in 9 mL of MRS Broth respectively then incubated at 10 °C, 37 °C, and 45 °C for 24 hours. The observation of LAB growth on different temperature in 3 equal replicates. The positive result or growth was shown from turbidity formation. This turbidity showed that the LAB had ability to grow on the assay temperature.

If there was no LAB growth, the medium was still clear or similar with MRS Broth medium (Harrigan and Cance, 1976). The optical density of cell was measured using UV-VIS spectrophotometer at  $\lambda_{600\text{ nm}}$ . Data of absorbance value was input into the quation of standard curve to determine the population of LAB at different incubation temperature.

#### **Experimental Design**

A factorial completely randomized design was performed to determine the population of LAB at certain temperature with different treatment factor of temperature as follows, P1:10 °C, P2:37 °C, and P3:45 °C and also the different type of isolate. Each treatments was repeated 3 times.

#### **Data Analysis**

The data was analysed to obtained average value and deviation standard of LAB growth. The differentiation on calculation of bacteria population at different temeperature treatments (10 °C, 37 °C, and 45 °C) and different isolate type (A dan B) was analysed using ANOVA Tukey test was done to verify if there is a significant difference.(Mattjik dan Sumertajaya 2013).

## Results and Discussion

### The pH and TAT Value of Dangke

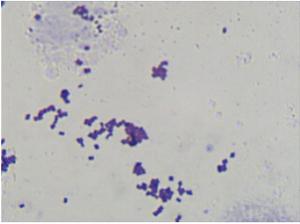
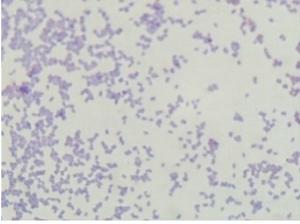
This study showed that the pH value of dangke was  $5.22 \pm 0.02$ . The result was similar with Aras (2009) reported that dangke had the pH value about 6. Hatta *et al* (2013) also reported that the pH value of dangke made from buffalo and bovine milk had no different around 6.4. Dangke was one of Indonesian native dairy product which had been known as traditional cheese by consumers (Ingrid *et al.*, 2000). This product had been processed in some districts in South Sulawesi (Enrekang, Sinjai, Anggeraja, and Alla). Marzoeki *et al.* (1978) stated that the physical characteristic of dangke with good quality was white and elastic. Nielsen (2003) stated that pH value reflected the concentration of free hydrogen ions which contained in a product. The pH value determined the acidity of the product, therefore if the pH value in product was low then the acidity level was high and the other way, so it could be stated that the level of acidity in dangke in this study was high.

Nielsen (2003) reported that total lactic acid of food could be measured through the Total Titratable Acid (TAT) test. If the pH value of a product decreased, the value of total lactic acid increased. The TAT value of dangke in this study was  $1.28 \pm 0.34\%$ . Syah (2012) reported that the average value of TAT of dangke was 0.17% to 0.63%, the concentration treatments of lactic acid bacteria, temperature, and different storage time affected the total value of lactic acid in dangke.

### Isolation and Identification of LAB

Twenty four isolates were successfully isolated from dangke. Furthermore, all isolates were identified microbiologically based on observation of cell morphology, catalase test, and Gram staining. Identification result shown in Table 1, it showed that only two isolates identified as LAB (DK2a1 and DK3a12). Characteristics of both isolates were cocci shape-cell that formed tetrad, negative catalase (there were no gasses formed), and Gram-positive (purple cells wall). According to Fardiaz (1989) LAB had same characteristic similar with these isolates, which had cocci shape-cell or basil, Gram-positive, negative catalase, and no spores.

Table 1. Microbiology Identification of LAB from Dangke

Isolate Code	Morphology	Catalase	Gram Staining
DK 2a1	 Cocci	Negative	Gram-positive
DK 3a12	 Cocci	Negative	Gram-positive

The ability of isolates to ferment carbohydrates characterized by a positive sign that changed the color of CHL medium from red to yellow and the pH values was acidic (pH 2-5). Based on the Api Web analysis (API 50CHLV5.0), the lactic LAB isolates of dangke DK 2a1 and 3a12 was identified as *Pediococcus pentosaceus* MSS 1 and *Pediococcus pentosaceus* MSS by 99.9% of similarity. Both isolates had the same ability to ferment carbohydrates for 48 hours of incubation. Some types of monosaccharides carbohydrates capable to fermented such as L-arabinose, D-Ribose, D-Galactose, D-Glucose, D-Fructose, D-Mannose, and other carbohydrates such as N-acetylglucosamine, Amygladin, arbutin, esculin, ferrocitrate, salicin, D-Celiobiose, Maltose, D-Trehalose, Gentiobiose, and D-Tagatose.

Furthermore, the isolates were identified biochemically based on their ability to ferment carbohydrate using API 50 CHL kit to determine the species of LAB from dangke. LAB identification result using kit API 50 CHL was shown in Table 2.

Table 2. LAB Identification of dangke isolates using API 50 CHL kit

Tube	Carbohydrate	Isolate DK 2a1*		pH	Isolate DK 3a12**		pH
		24h	48 h		24h	48h	
0	Control	-	-	7	-	-	7
1	Glycerol	-	-	7	-	-	6
2	Erythritol	-	-	7	-	-	6
3	D-Arabinose	-	-	7	-	-	6
4	L-Arabinose	-	+	5	-	+	5
5	D-Ribose	+	+	5	+	+	5
6	D-Xylose	-	-	6	-	-	6
7	L-Xylose	-	-	6	-	-	7
8	D-Adonitol	-	-	7	-	-	7
9	Methyl-βD-Xylopyranoside	-	-	7	-	-	7
10	D-Galactose	+	+	4	+	+	4
11	D-Glucose	+	+	4	+	+	4
12	D-Fructose	+	+	4	+	+	4
13	D-Mannose	+	+	4	+	+	4
14	L-Sorbose	-	-	6	-	-	6
15	L-Rhamnose	-	-	6	-	-	7
16	Dulcitol	-	-	6	-	-	6
17	Inucitol	-	-	6	-	-	7
18	D-Mannitol	-	-	6	-	-	7
19	D-Sorbitol	-	-	6	-	-	7
20	Methyl-αD-Mannopyranose	-	-	6	-	-	7
21	Methyl-αD-Glucopyranose	-	-	6	-	-	6
22	N-AcetylGlucosamine	+	+	5	+	+	5
23	Amygladin	+	+	5	+	+	5
24	Arbutin	+	+	5	+	+	5
25	Esculin feric citrate	+	+	13	+	+	13
26	Salicin	+	+	5	+	+	5
27	D-Celiobiose	+	+	4	+	+	4
28	Maltose	+	+	4	+	+	4
29	D-Lactose	-	-	6	-	-	6
30	D-Melibiose	-	-	6	-	-	7
31	D-Saccharose	-	-	6	-	-	7
32	D-Trehalose	+	+	4	+	+	4
33	Inulin	-	-	6	-	-	6
34	D-Melezitose	-	-	6	-	-	6
35	D-Rafinose	-	-	6	-	-	7
36	Amidon	-	-	7	-	-	7
37	Glycogen	-	-	7	-	-	7
38	Xylitol	-	-	7	-	-	6
39	Gentiobiose	+	+	4	+	+	5
40	D-Turanose	-	-	7	-	-	7
41	D-Lyxose	-	-	6	-	-	7
42	D-Tagatose	+	+	4	+	+	4
43	D-Fucose	-	-	6	-	-	7
44	L-Fucose	-	-	7	-	-	7
45	D-Arabitol	-	-	7	-	-	7
46	-Arabitol	-	-	7	-	-	7
47	Potassium Gluconate	-	-	7	-	-	7
48	Pottasium 2-Ketogluconate	-	-	8	-	-	8
49	Potassium 5-Ketogluconate	-	-	7	-	-	7

(+) : able to ferment; (-) : not able to ferment

\* *Pediococci pentosaceus* MSS 1; \*\**Pediococci pentosaceus* MSS 2

According to Ray (2003), *Pediococci* was cocci shaped-cell and formed tetrad cells, able to live in pairs, including gram-positive, non-motile, no spores, and classified as facultative anaerobic bacteria. Depend on the species, the bacteria could ferment sucrose, arabinose, ribose, and xylose. It was also consistent with the results of study conducted by Abbasiliasi *et al.* (2012), stated that the *Pediococci* strain were gram-positive, catalase negative, rounded form tetrads and had the ability to grow in salt conditions with inclusion of 2% NaCl concentration and temperature range 30°C to 45°C. Wikandari *et al.* (2012) also reported that *Pediococci* isolated from bekasam (fermented fish) had characteristics as followed: not produced gas, able to survive in the 6.5 % of salt content and the condition of pH 4.2 - 9.6. According to Kiran *et al.* (2012), *Pediococci* was LAB that included to homo fermentative facultative bacteria with the metabolite such as lactic acid that played an important role in food fermentation and generally used in the fermentation of vegetables and sausages in natural or controlled condition.

The study conducted by Razak *et al.*(2009) stated that the LAB isolation of dangke originated from Enrekang, South Sulawesi resulted 30 isolates and the 3 isolates produced antimicrobial compounds and each identified as *Lactobacillus plantarum* DU15, *Enterococcifaecium* DU55, and *Leuconostoc mesentroides* DU02. Bacterial antimicrobial activity test was conducted to assay the bacterial pathogen *Salmonella typhimurium* FNCC0050. The difference LAB species of dangke was suggested due to the differences in the type of milk which used and the origin of dangke preparation (Razak *et al.*, 2009). Samples of dangke on this study made from cow's milk originated from the region of Sinjai, South Sulawesi, while samples of dangke on of Razak *et al.*(2009) study made from buffalo milk and originated from Enrekang, South Sulawesi.

### Growth of *P. pentosaceus* MSS 1 and *P. pentosaceus* MSS 2 at 10°C, 37 °C, and 45 °C

The results in Table 3 showed that different incubation temperatures had significant effect ( $P < 0.05$ ) to the average population of *P. pentosaceus* MSS 1 and *P. pentosaceus* MSS 2 for 24 hours of incubation, but there was no interaction between incubation temperature with LAB species to the end LAB population of dangke ( $P > 0.05$ ). Rahayu and Nurwitri (2012) stated that the temperature played important role in the growth of bacteria. *Pediococcus pentosaceus* MSS 1 and *P. pentosaceus* MSS 2 that incubated at 10°C had growth inhibition and negative value of average percentage in population as seen in Figure 1. This result was different to the temperature treatments at 37°C and 45°C. At 37 °C of incubation, *P. pentosaceus* MSS 1 and *P. pentosaceus* MSS 2 had optimal growth with the average population of 9.78 log 10 cfu mL<sup>-1</sup> and 9.65 log 10 cfu mL<sup>-1</sup> and the average percentage enhancement in population was higher (11.86%) than the initial population (10.71%) (Figure 1).

Table 3. The average population of *P. pentosaceus* MSS1 and *P. pentosaceus* MSS2 at different temperatures

Species of LAB	P0 h (log 10 cfu mL <sup>-1</sup> )	Average P24h in temperature at		
		10 °C	37 °C	45 °C
<i>P. pentosaceus</i> MSS 1	8.83	8.33 ± 0.10 <sup>c</sup>	9.78 ± 0.10 <sup>a</sup>	9.59 ± 0.04 <sup>a</sup>
<i>P. pentosaceus</i> MSS 2	8.62	8.06 ± 0.03 <sup>d</sup>	9.65 ± 0.11 <sup>a</sup>	9.31 ± 0.11 <sup>b</sup>

Note: a,b,c,d : numbers in different column and row followed by different superscript showed significant different at 5 % level test (Tukey), P0: initial population at 0 hour of incubation, P24: end population at 24 hours of incubation.

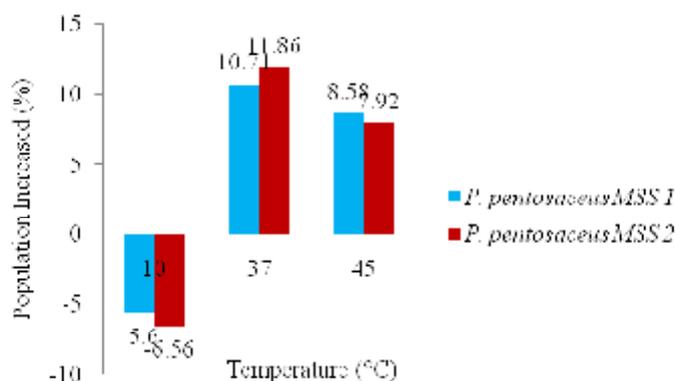


Figure 1. Relationship between incubation temperature with population enhancement of *P. pentosaceus* MSS 1 and *P. pentosaceus* MSS 2 from 0 up to 24 hours of incubation

At 45 °C of incubation, the average population of *P. pentosaceus* MSS1 and *P. pentosaceus* MSS2 were 9.59 log 10 cfu mL<sup>-1</sup> and 9.31 log 10 cfu mL<sup>-1</sup> with the average percentage enhancement in population 8.58 % and 7.92 % (Figure 1). It is concluded that the bacteria were able to survive at a fairly high temperature conditions, but not optimal.

Papagianni and Anastasiadou (2009) stated that the variations in the different species and strains produced differences intolerance to oxygen, pH, temperature, and NaCl and its ability to ferment carbohydrates.

## Conclusion

It could be concluded that the LAB of dangke originated from Sinjai, South Sulawesi were identified *P. pentosaceus* MSS 1 and *P. pentosaceus* MSS 2. This LAB had gram-positive characteristic, cocci shape-cell formed tetrad, negative catalase, homo fermentative and mesophilic bacteria that had optimum growth at 37°C and able to survive in high concentration of NaCl 6.5%. Both of this species could survive at 45 °C and the growth was inhibited at 10°C.

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# Bacteriological Quality of *Se'i* Treated with Liquid Smoke

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## Abstract

Using of liquid smoke in meat processing is widely used. However, the price of liquid smoke is quite expensive, thus it is good to combine with any kind of herbal to reduce production price as well as the quality meat processing products. One alternative herbal is roselle (*Hibiscus sabdariffa*). Effectiveness of *Schleichera oleosa*, *Zizyphus mauritiana lamk*, *Psidium guajava* and coconut shell liquid smoke on reducing bacteria count in *se'i* (Rotenese smoked meat) was investigated. Completely randomized design 5 x 3 was assigned in this experiment. The 5 treatments were: C= Smoked traditionally (without liquid smoke) as control, *Schleichera oleosa* (SOLS), *Zizyphus mauritiana lamk* (ZMLS), *Psidium guajava* (PGLS) and coconut shell liquid smoke (CSLS). Each treatment had 3 replications. Parameters measured were aroma, taste, total bacteria count, *Escherichia coli* and *Staphylococcus aureus*. All parameters were evaluated after one day, 5, 10, 15 and 20 days of storage at 4°C. Analysis of variance (ANOVA) was applied followed by least square means (LSD test). Result of this experiment indicated that bacteriological quality were significantly effected ( $P < 0.05$ ) by liquid smoke. Addition of SOLS in *se'i* could inhibit growth rate of total bacteria and *Staphylococcus aureus* up to 20 days, whereas PGLS up to 15 days and, ZMLS, CSLS and control just up to 10 days of storage time. It indicates that SOLS is more effective to maintain bacteriological quality up to 20 days of storage time than PGLS, ZMLS and CLLS.

**Keywords:** bacteria count, liquid smoke, *se'i*,

## Introduction

Application of liquid smoke has been widely used in food industry including: tofu, noodle, meat and fish. It was reported that application of liquid smoke in food could deteriorate some kinds of bacteria such as: *Escherichia coli*, *Staphylococcus aureus*, *Bacillus subtilis*, *Pseudomonas fluorence* (Yulistiana *et al.*, 1997), *Salmonella* (Putnam *et al.*, 1999), total bacteria number (Putranto *et al.*, 2009) and *Pseudomonas aeruginosa* (Zuraida *et al.*, 2011) so it could prolong self life of the food.

Tars and polycyclic aromatic hydrocarbons (PAH) which are recognized as being carcinogens and always found in gas smoke, were not found in liquid smoke (Budijanto *et al.*, 2008). Thus liquid smoke is save to be applied in food including *se'i* (Rotenese smoked meat). *Se'i* is always processed traditionally used Kusambi (*Schleichera oleosa*) wood as a fuel. But sometimes *Zizyphus mauritiana lamk*, *Psidium guajava* wood and coconut shell always used as a fuel in *se'i* processing. The objectives of this study were to evaluate the ability of four kinds of liquid smoke: *Schleichera oleosa*, *Zizyphus mauritiana lamk* and *Psidium guajava* to inhibit the growth of total bacteria, Coliform, *Escherichia coli* and *Staphylococcus aureus* in *se'i*.

## Materials and Methods

Eighteenth Kgs of beef obtain from rump were bought in meat shop in Kupang. Salt, saltpeter ( $KNO_3$ ) and three kind of wood: *Schleichera oleosa*, *Zizyphus mauritiana lamk*, *Psidium guajava*, and coconut shell were purchased from a local market in Kupang. All woods and coconut shell cut 3 x 3 cm in size for making liquid smoke at 400°C pyrolysis temperature. The liquid smoke was produced in Department of Education, Chemistry division Nusa Cendana University, Indonesia.

## *Se'i* Processing

The beef were trimmed off the excessive fat, sliced in rope-shape (*lalolak*), added 500 mgs of saltpeter and 2% of salt  $kg^{-1}$  of beef, mixed well then split into 5 batches (saw detail in experimental design) to which the liquid smoke was added separately. The liquid smoke then injected to meat 1 mL  $kg^{-1}$  at 12-15 different

part of sliced beef, then marinated  $\pm$  12 hours. After marinating, each batch was smoked separately. The control was smoked using *Schleichera oleosa* wood while the batches which applying the liquid smoke were smoked using Hock stove (22 wicks). After smoking each batch was packaged separately in polyethylene bags and chilled for 0, 5, 10, 15 and 20 days at 4°C.

### Experimental Design, Parameters Measured and Statistical Analysis

Design under experiment was completely randomized design (CRD). The five treatments consisted of: smoked traditionally (without liquid smoke) as control (C), *Schleichera oleosa* liquid smoke (SOLS) 1% (v/w), *Zizyphus mauritiana lamk liquid smoke* (ZMLS) 1% (v/w), *Psidium guajava liquid smoke* (PGLS) 1% (v/w) and *coconut shell liquid smoke* (CSLS) 1% (v/w). For each treatment, measurements were made in triplicate.

Variables studied were total bacterial count, Coliform, *Escherichia coli* and *Staphylococcus aureus* which followed procedure describe by Harrigan and McCance (1976). Bacterial data was analyzed using analysis of variance (ANOVA). To determine where there were significant differences between the treatment, least square difference (LSD) between means were made (SPSS 18). The analysis was performed one day after processing and after 5, 10, 15 and 20 days of chilling storage.

### Results and Discussion

Maximum level of total bacteria count permitted in smoke meat is  $1 \times 10^5$  CFU, Coliform 10 mpn or 1.0 log MPN/g and *Staphylococcus aureus* is negative (National Standard Board of Indonesia, 2009). It could be noticed in Table I that as the storage time increased the total bacteria count of *se'i* samples also increased gradually ( $P < 0.05$ ), with the lowest rates was in SOLS samples compared to others samples. At the end of storage time (20 days) the bacterial count of control, SOLS, ZMLS, PGLS and CSLS reached the highest number but just *se'i* samples treated with SOLS had the number of bacteria below  $1 \times 10^5$  CFU or 5 log CFU/g. All *se'i* samples were good to be consumed up to 15 days of storage time, and only *se'i* treated with SOLS could be consumed up to 20 days of storage time.

Table 1. Total bacterial, Coliform and *Staphylococcus aureus* of *Se'i* treated with liquid smoke during storage time

Liquid smoke	Storage time (days)				
	0	5	10	15	20
Total bacterial count (log CFU/ g)					
C	10 <sup>aA</sup>	2.56 $\pm$ 0.01 <sup>bA</sup>	3.63 $\pm$ 0.03 <sup>cA</sup>	4.76 $\pm$ 0.01 <sup>dA</sup>	6.13 $\pm$ 0.02 <sup>eA</sup>
SOLS	10 <sup>aA</sup>	1.79 $\pm$ 0.03 <sup>bB</sup>	1.66 $\pm$ 0.16 <sup>bB</sup>	2.09 $\pm$ 0.12 <sup>cB</sup>	4.45 $\pm$ 0.12 <sup>dB</sup>
ZMLS	10 <sup>aA</sup>	2.50 $\pm$ 0.31 <sup>bA</sup>	3.17 $\pm$ 0.02 <sup>cA</sup>	4.80 $\pm$ 0.06 <sup>dA</sup>	5.27 $\pm$ 0.01 <sup>eC</sup>
PGLS	10 <sup>aA</sup>	2.37 $\pm$ 0.05 <sup>bA</sup>	2.71 $\pm$ 0.23 <sup>bC</sup>	3.86 $\pm$ 0.01 <sup>cC</sup>	5.14 $\pm$ 0.13 <sup>dC</sup>
CSLS	10 <sup>aA</sup>	2.42 $\pm$ 0.11 <sup>bA</sup>	3.27 $\pm$ 0.01 <sup>bA</sup>	4.37 $\pm$ 0.13 <sup>cC</sup>	5.41 $\pm$ 0.01 <sup>dC</sup>
Coliform (log MPN/ g)					
C	0 <sup>aA</sup>	0 <sup>aA</sup>	1.0 $\pm$ 0.23 <sup>aA</sup>	2.65 $\pm$ 0.02 <sup>bA</sup>	3.35 $\pm$ 0.12 <sup>cA</sup>
SOLS	0 <sup>aA</sup>	0 <sup>aA</sup>	1.0 $\pm$ 0.21 <sup>aA</sup>	1.0 $\pm$ 0.03 <sup>bB</sup>	1.0 $\pm$ 0.11 <sup>bB</sup>
ZMLS	0 <sup>aA</sup>	0 <sup>aA</sup>	1.0 $\pm$ 0.11 <sup>aA</sup>	2.51 $\pm$ 0.01 <sup>bA</sup>	3.29 $\pm$ 0.17 <sup>cC</sup>
PGLS	0 <sup>aA</sup>	0 <sup>aA</sup>	1.0 $\pm$ 0.02 <sup>aA</sup>	1.0 $\pm$ 0.01 <sup>bB</sup>	2.84 $\pm$ 0.23 <sup>cA</sup>
CSLS	0 <sup>aA</sup>	0 <sup>aA</sup>	1.0 $\pm$ 0.02 <sup>aA</sup>	2.48 $\pm$ 0.03 <sup>bA</sup>	2.84 $\pm$ 0.23 <sup>cA</sup>
<i>Staphylococcus aureus</i> (log MPN/g)					
C	0 <sup>aA</sup>	0 <sup>aA</sup>	0 <sup>aA</sup>	0 <sup>aA</sup>	1.0 $\pm$ 0.13 <sup>aA</sup>
SOLS	0 <sup>aA</sup>	0 <sup>aA</sup>	0 <sup>aA</sup>	0 <sup>aA</sup>	1.0 $\pm$ 0.03 <sup>aA</sup>
ZMLS	0 <sup>aA</sup>	0 <sup>aA</sup>	0 <sup>aA</sup>	2.92 $\pm$ 0.13 <sup>cA bB</sup>	3.73 $\pm$ 0.21 <sup>bB</sup>
PGLS	0 <sup>aA</sup>	0 <sup>aA</sup>	0 <sup>aA</sup>	0 <sup>aA</sup>	1.0 $\pm$ 0.33 <sup>aA</sup>
CSLS	0 <sup>aA</sup>	0 <sup>aA</sup>	0 <sup>aA</sup>	2.85 $\pm$ 0.11 <sup>aB</sup>	3.58 $\pm$ 0.03 <sup>bB</sup>

<sup>a,b,c</sup> Means with different small letter within same row are significantly different ( $P < 0.05$ )

<sup>A,B,C</sup> Means with different capital letter within same column are significantly different ( $P < 0.05$ )

CFU: colony forming unit. MPN: most probably number.  $\pm$  standard deviation (SD)

Coliform count in control (C), ZMLS and CSLS *se'i* samples were in good quality up to 10 days, while PGLS and SOLS were still in good quality up to 15 days and 20 days of storage time respectively. Thus *Schleicheria oleosa* liquid smoke was the best treatment to inhibit coliform growth during storage. Based on the coliform data, all *se'i* samples were good to be consumed up to 10 days of storage time, and only *se'i* treated with SOLS could be consumed up to 20 days of storage time.

*Staphylococcus aureus* is a pathogen bacterial and used as bacteria indicator of safety food especially for processing food such as smoked meat. From Table 1, apparent that up to 10 days of storage time *Staphylococcus aureus* was not detected in all *se'i* samples, however, at 15 days of storage time *Staphylococcus aureus* begun to be detected at ZMLS and CSLS *se'i* sample and at the end of storage time (20 days) all *se'i* samples were detected ( $P < 0.05$ ). All *se'i* sample free of *Escherichia coli*. This data showed that all *se'i* samples were good to be consumed up to 10 days of storage time, and only *se'i* treated with SOLS could be consumed up to 20 days of storage time. Based on the National Standard Board of Indonesia, this result indicated that all *se'i* samples were good to be consumed up to 10 days of chilling storage time, and only *se'i* treated with SOLS could be consumed up to 20 days.

## Conclusion

*Schleicheria oleosa* liquid smoke appear to be the most effective liquid smoke in controlling growth of total bacteria, Coliform, *Staphylococcus aureus* and *Escherichia coli* during the storage of *se'i* compare to other liquid smoke, followed by *Psidium guajava* liquid smoke.

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# Increase on Commercial Weight, Carcass Quality and Economic Benefit of Selected Local Meat Chicken Fed on Fermented Diet Contained Digestive Enzymes and Probiotics

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## Abstract

Three hundred (300) male local meat selected chicken were used to investigate the growth, commercial weight, carcass quality and economic benefit of local meat selected chicken fed on with fermented diet based on local material contained, digestive enzymes and probiotic. The aim of present study was to continue the genetic development program of local chicken by management and nutritional approach. Research was carried out for 12 month on 3 generation of offspring which were F8, F9 and F10 of selected local meat chicken. Chicken were divided in 3 group of body weight; light, medium and heavy type after hatching. The fermented diet as treatment was composed by rice bran, corn bran, palm kernel and sago waste. Complete digestive enzymes and probiotic were used as feed additive. After 1 week of age, chicken were fed on research diet which contained 0, 10, 20 and 30% of fermented diet and commercial diet for meat chicken. Observation was carried out for 90 days on each generation of local selected meat chicken. Research parameters were; average body weight, commercial weight, feed conversion and efficiency carcass composition, carcass quality especially protein, fat and cholesterol contents. The economic benefit was also analyzed by BC ratio. The results showed that supplementation of fermented diet composed by local materials and contained digestive enzyme and probiotic by as much as 10-30% of total diet has a similar effect on chicken fed on 100% of commercial diet. This phenomenon was observed on the growth, feed conversion and efficiency as well as the carcass composition and carcass quality. The commercial weight on 90 days of age was 1,27 kg (light type), 1,31 kg (medium type) and 1,47 kg (heavy type) on chicken fed on 10, 20 and 30% of fermented diet, respectively. Chicken type also showed the differences on carcass weight especially on breast and thigh muscles. This present study also showed very interesting where the supplementation of fermented diet contained a complete digestive enzymes and probiotics increased carcass percentage and carcass composition but it reduced fat and cholesterol content of the local broiler chicken meat in each generation. Economically, supplementation of 10-30% fermented diet also reduced feed cost between 16- 21% in the medium and heavy type of chicken and BC ratio was 1.31 to 1.37. In conclusion, supplementation of fermented diet containing a complete digestive enzymes and probiotics suppressed the use of commercial feed until 30% and provided advantages in terms of production, carcass quality and economic benefit of the selected local meat chicken.

Keywords: local chicken, fermented diet, digestive enzymes, probiotic.

## Introduction

In the business of local poultry, feeding quality is the main key which must be arranged as a part of operational management to increase profits. Such efforts can be done by using genetic quality of DOC through selection and breeding program which is accompanied by the use of local materials and feed additives for feeding formulation. Historically, genetic selection process has produced a kind of selected local chicken that has a better growth rate than its origin and the utilization of local materials as poultry feeding mixture with various digestive enzyme and probiotic as result of animal feed technology has been proven reducing the feed cost, stimulate rate of growth and carcass quality, and increase business profits (Yaman, 2010:2013). However, the use of local raw materials still have a problem because the majority resulted by agricultural and industrial waste that has characteristics of high crude fiber and low protein. Fermentation, supplementation of digestive enzyme and probiotics in animal feeding is a strategy to improve the biological and nutritional values of local materials as feeding materials of local meat chicken. Local materials such as rice bran, corn pulp, sago waste and tofu waste are materials that can be used as nutrient source of fermented feed for poultry in order to suppress the use of commercial feeding.

Digestive enzymes are protein molecules that catalyze specific chemical reactions during digestive process. In most cases, feeding enzymes is believed to increase the rate and extent of digestion of certain nutrients for chicken production (Beedford, 2000). A number of enzyme products and probiotics are currently on the market intended for addition to poultry feed. Most of these are digestive enzymes and probiotics, intended to improve the chicken's ability to digest nutrients in the feed. It was clear that the combination of digestive enzyme and probiotics in fermented diet of local meat chicken is expected to increase the feed efficiency and digestibility (Khattack *et al.*, 2006).

This study aims to improve the nutritional benefits of local materials as feed supplements of meat local chicken to improve the production and carcass quality of local meat chicken carcass to provide maximum benefit to the farmers. Although the use of digestive enzyme and probiotics to improve the nutritional value of raw materials for chicken feeding is common practice, the application on fermented feeding is not as widespread. Despite these challenges, the application of fermented feeding combine with digestive enzyme and probiotics to achieve nutritional goals and profits within the local meat chicken business is likely to continue well into the future in Indonesia.

## Materials and Methods

The present study was designed by a randomized factorial design, using a 3-generation of offspring, 3 growth types and 4 treatments of feeding. Each treatment consisted of 5 replications and each replication consisted of 5 chicks of local meat chickens. Research was carried out for 12 month on 3 generation of offspring which were F8, F9 and F10 of selected local meat chicken. Chicken were divided in 3 group of body weight; light, medium and heavy type after hatching.

The fermented diet as treatment was composed by rice bran, corn bran, palm kernel, tofu and sago waste. Complete digestive enzymes and probiotic were used as feed additive. After 1 week of age, chicken were fed on research diet which contained 0, 10, 20 and 30% of fermented diet and commercial diet for meat chicken. Observation was carried out for 90 days on each generation of local selected meat chicken.

Research parameters were; average body weight, commercial weight, feed conversion and efficiency carcass composition, carcass quality especially protein, fat and cholesterol contents. The economic benefit was also analyzed by BC ratio.

Table 1. The formulation of feeding treatments on selected local meat chicken fed on fermented diet contained digestive enzymes and probiotics

Materials	Combination of feeding (%)			
	A	B	C	D
Commercial diet	100	90	80	30
Feeding treatments	0	10	20	70
Protein content	21.02	21.08	21.13	21.21
Metabolism energy (kcal/kg)	3010	3015	30124	3048

## Results and Discussion

The present results showed that there was a different response between generations and types of local meat chickens to the percentage of feed. In this study, the tenth generation of local meat chickens has a better response than the eighth and ninth generations on nutritional factors. The results also showed that supplementation of fermented diet composed by local materials and contained digestive enzyme and probiotic by as much as 10-30% of total diet has a better effect on chicken fed on 100% of commercial diet. This phenomenon was observed on the growth, feed conversion and efficiency as well as the carcass composition and carcass quality. The commercial weight on 90 days of age was 1,27 kg (light type), 1,31 kg (medium type) and 1,47 kg (heavy type) on chicken fed on 10, 20 and 30% of fermented diet, respectively. Chicken type also showed the differences on carcass weight especially on breast and thigh muscles.

This present study also showed very interesting information where the supplementation of fermented diet contained a complete digestive enzyme and probiotics increased carcass percentage and carcass composition but it reduced fat and cholesterol content of the local broiler chicken meat in each generation.

Table 2. Performance and carcass percentage of selected local meat chicken fed on fermented diet contained digestive enzymes and probiotics

Parameters	Chicken Type	Combination of feeding (%)			
		A	B	C	D
DOC weight (gram)	light	29.3	29.3	29.6	29.4
	medium	34.2	34.1	34.3	34.3
	heavy	37.6	37.5	37.5	37.5
Final body weight (gram)	light	989.5	988.9	999.1	989.9
	medium	1022.4 <sup>a</sup>	1041.6 <sup>a</sup>	1247.3 <sup>b</sup>	1277.2 <sup>b</sup>
	heavy	1155.7 <sup>a</sup>	1272.4 <sup>b</sup>	1315.2 <sup>b</sup>	1472.5 <sup>b</sup>
Carcass weight (gram)	light	682.8 <sup>a</sup>	702.1 <sup>a</sup>	762.3 <sup>b</sup>	772.1 <sup>b</sup>
	medium	736.1 <sup>a</sup>	770.8 <sup>a</sup>	947.9 <sup>b</sup>	957.9 <sup>b</sup>
	heavy	820.5 <sup>a</sup>	928.9 <sup>b</sup>	999.6 <sup>b</sup>	1,133.8 <sup>c</sup>

Means in the same row with different superscript differ significantly (p<0.05).

In terms of carcass quality, an increasing of protein content and decreasing of fat and cholesterol in local meat chicken has a close correlation on the increase of fermented diet contained digestive enzymes and probiotics. Improvement of chicken body weight and carcass quality of the local meat chicken fed on fermented feeding contained digestive enzymes and probiotics due to increase on feed quality and feed digestibility (Cowieson and Adeola, 2005). It was clear that digestive enzymes and probiotics are parts of feed additive stimulate the feed efficiency and nutritive value for poultry (Kocher *et al.*, 2002).

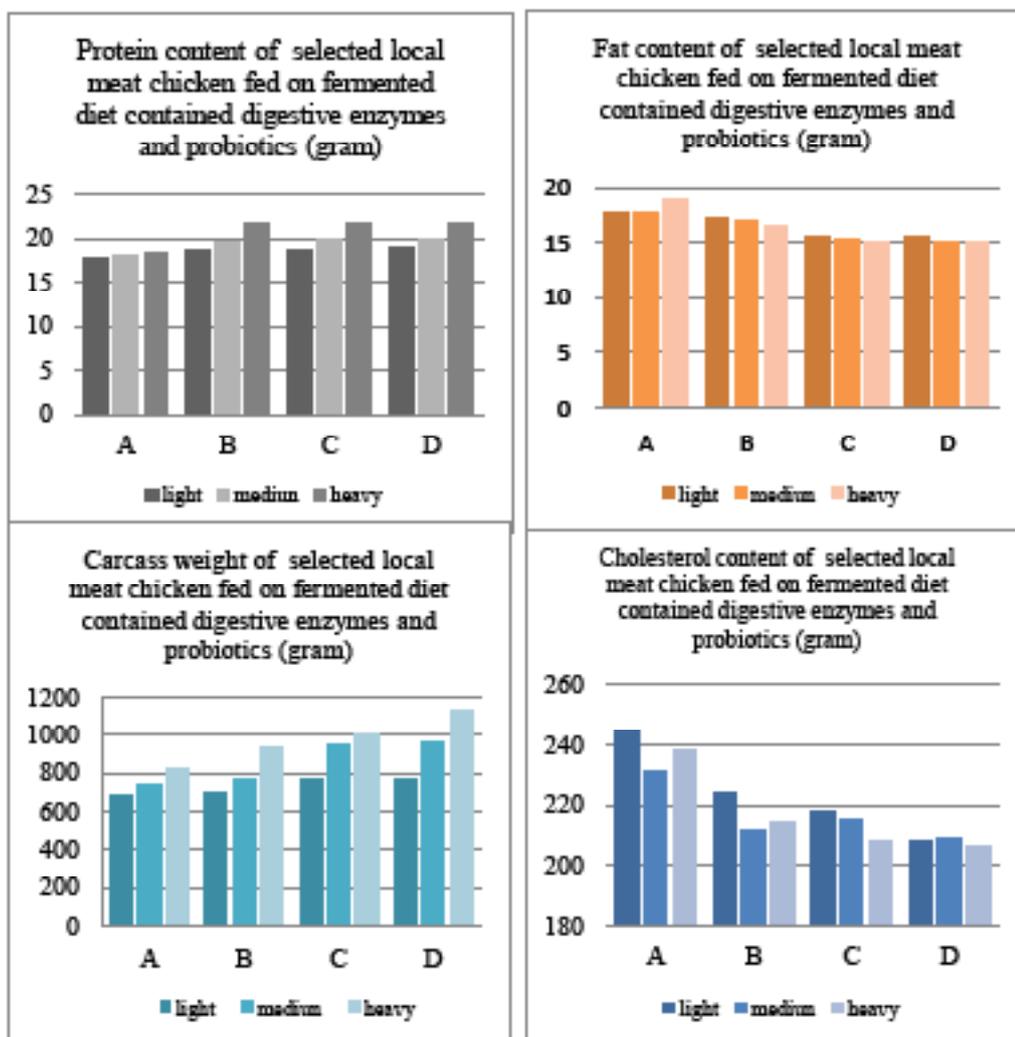
Table 3. Performance and carcass percentage of selected local meat chicken fed on fermented diet contained digestive enzymes and probiotics

Parameters	Chicken Type	Combination of feeding (%)			
		A	B	C	D
Protein (%)	light	17.9	18.7	18.7	19.0
	medium	18.2	19.7	19.9	19.8
	heavy	18.4 <sup>a</sup>	21.6 <sup>b</sup>	21.8 <sup>b</sup>	21.6 <sup>b</sup>
Fat (%/BK)	light	17.8 <sup>a</sup>	17.2 <sup>a</sup>	15.6 <sup>b</sup>	15.4 <sup>b</sup>
	medium	17.7 <sup>a</sup>	16.9 <sup>a</sup>	15.2 <sup>b</sup>	15.1 <sup>b</sup>
	heavy	18.8 <sup>a</sup>	16.4 <sup>b</sup>	15.1 <sup>b</sup>	15.1 <sup>b</sup>
Cholesterol (mg/100gr)	light	244.6 <sup>a</sup>	223.8 <sup>a</sup>	217.6 <sup>b</sup>	207.8 <sup>b</sup>
	medium	231.3 <sup>a</sup>	211.4 <sup>b</sup>	215.2 <sup>b</sup>	208.9 <sup>b</sup>
	heavy	237.8 <sup>a</sup>	214.2 <sup>b</sup>	207.6 <sup>b</sup>	206.5 <sup>b</sup>

Means in the same row with different superscript differ significantly (p<0.05).

Some of the digestive enzymes have potential for use in the feed industry include cellulase ( $\beta$ -glucanases), xylanases and associated enzymes, phytases, proteases, lipases, and galactosidases. Digestive enzymes have mostly been used for poultry to neutralize the effects of the viscous, nonstarch polysaccharides in cereals such as barley, wheat, rye, and triticale. These antinutritive carbohydrates are undesirable, as they reduce digestion and absorption of all nutrients in the diet, especially fat and protein (Odetallah, 2002.; Khattack *et al.*, 2006; Wang *et al.*, 2005). In addition, digestive enzymes and probiotics were known reduced beak impaction and vent plugging, decreased size of gastrointestinal tract, altered population of microorganisms in gastrointestinal tract, reduced water intake, reduced water content of excreta, reduced production of ammonia from excreta, reduced output of excreta, including reduced N and P (Lan *et al.*, 2003; Kabir *et al.*, 2004).

Economically, supplementation of 10-30% fermented diet also reduced feed cost between 16- 21% in the medium and heavy type of chicken and BC ratio was 1.31 to 1.37, In conclusion, supplementation of fermented diet containing a complete digestive enzymes and probiotics suppressed the use of commercial feed until 30% and provided advantages in terms of production, carcass quality and economic benefit of the selected local meat chicken. Results of this study also proved that medium and heavy type of selected local meat chicken have the ability to adapt to changes in the type and variety of feed was higher than light type.



## Summary

Results of this present study clearly indicated that substitution of local materials in order to reduce the use of expensive commercial feeding in business of local meaty chicken will be very advantageous when using fermented feeding, digestive enzyme and probiotic. This study was also able to prove that the combination of feed fermentation, digestive enzyme and probiotic not only stimulate growth process but was able to clearly improve the meat quality of local chicken through an increase in the amount of meat protein and suppress the amount of fat and cholesterol contents.

From an economic standpoint, the use of fermented feeding, digestive enzyme and probiotics has been able to provide benefits greater than the commercial feeding. This was due to fermented feeding, digestive enzyme and probiotics able to stimulate growth process, meat quality through an increase in digestibility and feed efficiency of local meat chicken.

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# An Analysis of Cattle Traders Practices on Animal Traceability in Malaysia

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## Abstract

*Animal traceability systems are commonly used to provide a means for ensuring product traceability, respond to infectious disease incursions (for example foot-and-mouth disease) and to support various aspects of herd management including monitoring of reproductive performance, assessment of artificial insemination schemes and verification of ownership. In 2009, Malaysia implemented a traceability system using radio frequency identification (RFID) tags. In addition to RFID tags, animals are typically identified using visual ear tags, rumen boluses and/or branding. This was a cross-sectional study of cattle traders in the Malaysian Peninsular. Our objectives were to evaluate cattle trader compliance with traceability regulations. Data were obtained from 190 questionnaires completed by cattle traders in 10 states in the Peninsula. Almost 70% of the respondents claimed that they complied with traceability regulations. Most respondents (85%) reported that they routinely tagged their cattle and up to 98% routinely applied for movement or slaughter permits prior to moving cattle from one location to another. Level of education, length of time in the cattle industry, the number of cattle reared or traded, types of husbandry system and attendance at courses or training on cattle farming were significantly associated with self-declared traceability compliance. We conclude that education and awareness programmes on cattle traceability should be carried out to increase the level of understanding about the importance of traceability system among Malaysian cattle traders.*

**Keywords:** animal production, cattle, Malaysia, practices, traceability

## Introduction

Animal identification and animal traceability systems are commonly used to provide a means for ensuring product traceability and to respond to infectious disease incursions (for example foot-and-mouth disease). The World Organisation for Animal Health (OIE) has defined the aspects of animal husbandry as part of the elements to be included in the design and implementation of an animal identification system to achieve animal traceability (OIE, 2014). In Malaysia, cattle are typically identified using visual ear tags, rumen boluses, tattoos and/or branding. In 2009, the government implemented a cattle traceability system using radio frequency identification (RFID) tags (Salina and Azmie, 2013). In this system, all cattle must have some form of permanent identification (either visual or RFID tags) prior to their being moved either inter- or intra-state (Azmi and Salina, 2009).

Cattle traders are a sub-group of Malaysian cattle producers that are regularly involved in importing, trading and slaughtering cattle. As prescribed by the Malaysian cattle traceability system, individuals carrying out these activities need to bear the cost of RFID tags. In addition, they need to apply for movement and slaughter permits to move cattle from one location to another. The user compliance with the system has not been evaluated. This paper describes a cross-sectional study on cattle traders in the Malaysian Peninsular with the objective of evaluating their compliance with the national traceability regulations.

## Materials and Methods

A questionnaire was designed to assess cattle trader practices on livestock traceability. Ten state veterinary offices were approached and agreed to participate in this study. The questionnaire was administered to 190 respondents selected randomly among traders applying for movement or slaughter permits from the state veterinary offices or during herd health visits. Information on social and demography, experience in the cattle industry and compliance with the Malaysian cattle traceability system were collected. Responses from completed questionnaires were entered and analysed using SPSS version 20 (IBM Corporation, 2011).

Chi-squared analyses were used to test the significance of the association between the socio-demographic and cattle industry background of traders, and the outcome of interest (defined as self-declared compliance to the cattle traceability system). Univariable logistic regression was used to quantify the strength of association between socio-demography and cattle industry background with the traceability compliance.

## Results and Discussion

Of the 190 traders participated in this study, 95% were males and the rest were females. Half of them were between 20 and 40 years of age at the time of study. More than half (67%) of respondents had at least a secondary education and 60% traded between 11 and 100 head of cattle at any one time. Most of the respondents (76%) also reared and bred cattle, and 48% practiced an intensive system. Almost three quarters (73%) of respondents reported that they had attended a course, seminar or training related to cattle farming.

Table 1. Responses of respondents to questions related to practices on traceability

Question	Yes <i>n</i> (%)	No <i>n</i> (%)
Was your cattle given an animal identification?	161 (84.7)	29 (15.3)
If yes, did you purchase the animal identification yourself?	118(73.3)	43(26.7)
Have you ever purchase/sell cattle?	190 (100.0)	0 (0.0)
Did you apply for movement/slaughter permit?	186 (97.9)	4 (2.1)
If yes, was the movement inter-state?	135 (71.1)	51 (26.8)
Do you keep record of sell and purchase of cattle?	132 (69.5)	58 (30.5)
Do you keep record of breeding/calving/death?	95 (50.0)	95 (50.0)

The proportion of responses based on questions related to practices of traceability was shown in Table 1. Among traders, 73% purchased the animal identification themselves. Traders were experienced in selling and purchasing cattle and 98% of them routinely applied for movement or slaughter permits. However, only 50-70% of them kept records of cattle they had purchased and sold, cattle they had bred and cattle that had died while under their care. These findings are consistent with the study by Adesokan and Ocheja (2014) who found that the majority of the livestock traders in Nigeria used tags or tattoo to identify their animals, however, they had poor practices of traceability in other areas.

Level of education, the number of years working in the cattle industry, the number of cattle reared or traded, the type of husbandry system practiced and whether or not respondents had attended courses or training on cattle farming were significantly associated with cattle traceability compliance (Table 2). Traders with a higher education level and had more experience in the cattle industry were more likely to practice good traceability which is in agreement with the findings of Oladele (2008) who reported that the farmers with a higher level of education and with longer experience in the agricultural sector were more willing to pay for extension services in Nigeria.

Traders who reared or traded between 11 and 1000 head of cattle were more likely to be cattle traceability compliant compared with those that traded less than 10 head of cattle and those that traded more than 1000 head of cattle. As reported by Adesokan and Ocheja (2014), the proportion of livestock traders with good practice of traceability increased with increasing number of animals they sent for slaughter every week. Traders who were involved in the intensive or feedlot cattle farming systems were more likely to be cattle traceability compliant.

Traders who never attended courses or training on cattle farming were less likely to be cattle traceability compliant. This was reflected by the high percentage of the respondents who reported that they had never attended either a course or training session on cattle farming. Lack of awareness on the importance of traceability systems could be the main reason for these findings.

Table 2. Univariable logistic regression analysis of factors affecting good traceability practice

Variables	Good <i>n</i> (%)	Poor <i>n</i> (%)	Total	OR <sup>a</sup>	95% CI <sup>b</sup>	<i>p</i> -value
Gender:						
Male	125 (69.4)	55 (30.6)	180	1	Reference	-
Female	7 (70.0)	3 (30.0)	10	1.03	0.26, 4.12	0.970
Age (years):						
20-40	71 (74.0)	25 (26.0)	96	1	Reference	-
41-60	49 (65.3)	26 (34.7)	75	0.66	0.34, 1.28	0.222
61 and above	12 (63.2)	7 (36.8)	19	0.60	0.21, 1.70	0.340
Level of education:						
No formal education	2 (33.3)	4 (66.7)	6	1	Reference	-
Primary	11 (42.3)	15 (57.7)	26	1.47	0.23, 9.49	0.688
Secondary	95 (74.2)	33 (25.8)	128	5.76	1.01, 32.90	0.049
Tertiary	24 (80.0)	6 (20.0)	30	8.00	1.17, 54.50	0.034
Length in cattle industry (years):						
Less than 5	12 (50.0)	12 (50.0)	24	1	Reference	-
5-10	42 (76.4)	13 (23.6)	55	3.23	1.17, 8.90	0.023
11-15	35 (76.1)	11 (23.9)	46	3.18	1.12, 9.08	0.030
16-20	21 (65.6)	11 (34.4)	32	1.91	0.65, 5.64	0.242
21 and above	22 (66.7)	11 (33.3)	33	2.00	1.17, 8.90	0.208
Number of cattle rear/trade (head):						
Less than 10	2 (18.2)	9 (81.8)	11	1	Reference	-
11-100	83 (72.8)	31 (27.2)	114	12.05	2.47, 58.89	0.002
101-1000	38 (80.9)	9 (19.1)	47	19.00	3.49, 103.56	0.001
1001 and above	9 (50.0)	9 (50.0)	18	4.50	0.75, 26.93	0.099
Type of husbandry system practiced:						
Extensive	16 (51.6)	15 (48.4)	31	1	Reference	-
Semi-intensive	32 (62.7)	19 (37.3)	51	1.58	0.64, 3.90	0.322
Intensive /Feedlot	71 (78.0)	20 (22.0)	91	3.33	1.41, 7.88	0.006
Integration	13 (76.5)	4 (23.5)	17	3.05	0.81, 11.45	0.099
Attend course/ training:						
Yes	106 (76.8)	32 (23.2)	138	1	Reference	-
No	26 (50.0)	26 (50.0)	52	0.30	0.15, 0.59	0.001

<sup>a</sup> OR: Odds ratio<sup>b</sup> CI: Confidence interval

## Conclusion

Overall, 70% of cattle traders claimed that they comply with the legislative requirements of Malaysia's cattle traceability system. Traders with a higher education level, had more experience in the cattle industry, reared or traded between 11 and 1000 head of cattle and practiced an intensive or feedlot system were more likely to be traceability compliant. Those who never attended courses or training on cattle farming were less likely to be traceability compliant. We conclude that education and awareness programs on cattle traceability should be carried out to increase the level of understanding about the importance of traceability system among Malaysian cattle traders.

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# Effect of Moisture Reduction Method, Storage Period and Temperature on Honey Quality

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## Abstract

High humidity in Indonesia causes high honey moisture therefore susceptible to fermentation. Honey moisture can be reduced through dehydration or dehumidification. Dehydration involves heating the honey, but not so in dehumidification. This difference affects honey quality, so does storage condition. This study was aimed at analyzing honey quality after it is dehydrated or dehumidified then stored at different period and temperature. Completely randomized and 2x2x2 factorial design were used in the first and second studies. The first study treatment was moisture reduction method/MRM (dehydration and dehumidification). The first, second, and third factors in the second study were MRM (dehydration and dehumidification), storage period (2 and 4 months), and storage temperature (3 and 28 °C). The data were analyzed by ANOVA and LSM at  $\alpha=0.05$ . The first study honey samples from both MRM were significantly different only in activity of diastase enzyme; dehydration lowering the activity, while dehumidification otherwise. The second study showed that honey moisture was affected by storage period (2 months < 4 months) and interaction between MRM and temperature (highest: dehydration and 28 °C); HMF by MRM (dehydration > dehumidification) and interaction between storage period and temperature (highest: 4 months at 28 °C); reducing sugar by MRM (dehydration < dehumidification) and storage period (2 < 4 months); diastase activity by storage period (2 < 4 months) and interaction between MRM and temperature (lowest: dehydration at 28 °C); acidity by storage temperature (3 < 28 °C) and period (2 < 4 months); yeast by MRM (dehydration < dehumidification), storage period (2 < 4 months) and temperature (3 < 28 °C). All honey samples stored at a temperature of 3 °C still met SNI honey standard.

Keywords: dehumidification, dehydration, honey, storage

## Introduction

Indonesian honey generally has a high moisture content, more than SNI 01-3545-2004 standard which set the maximum moisture content at 22% (BSN 1994); so it is susceptible to fermentation that can decrease the honey quality. Honey moisture content can be reduced by direct heating (boiled), indirect heating (dehydration), and evaporation through contact with dry air (dehumidification). Vacuum dehydrator is used in dehydration method, while dehumidifier in dehumidification method.

Honey heating, while decreases the moisture content, also kills yeast which causes fermentation (Papa *et al.*, 2009), but uncontrolled heating can damage the honey such as increases hydroxymethylfurfural (HMF) content and decreases activity of diastase enzyme (Ribeiro *et al.*, 2012; Sahinler and Gul, 2005). Reducing honey moisture content by evaporation indeed safer, but this method does not kill yeast. Yeast growth can be inhibited at low temperatures (Graham, 1992), and this is the reason to keep honey in refrigerator. This study is aimed at comparing the effect of moisture reduction method on honey storage quality in different storage temperature and period.

## Materials and Methods

Honey sample was 70kg kapok (*Ceiba pentandra* L.) honey which had been harvested 7 months prior to this study with 21.6% moisture content. Vacuum dehydrator with automatic timer and temperature control and dehumidifier with the ability to absorb 4.4 litre of vapor per hour were used in dehydration and dehumidification methods respectively. The study was done in 2 stages, (1) honey moisture reduction and (2) honey storage.

## Moisture reduction stage

Prior to moisture content reduction, the honey quality was analyzed. Its moisture content then reduced to 17% using 2 methods, namely dehydration and dehumidification. About 46 kg honey sample were heated in vacuum dehydrator at 57°C with the pressure of 1kg/cm<sup>2</sup> for an hour then the moisture content was measured. This process was done repeatedly until the moisture content reached 17%. For dehumidification method, 20 kg honey sample was poured into 8 plastic trays. The trays then were put in the dehumidification chamber which was set on Rh 45% to vaporized the moisture. The moisture content was checked everyday and dehumidification process would be ended when the moisture content reached 17%. After moisture reduction, honey from each methods were poured into 20 clear glass bottles as samples, and 4 bottles were analyzed for initial storage honey quality.

## Storage stage

Other 16 bottles honey samples from each moisture reduction method were randomly stored according to the treatments (Table 1). At the end of storage periods, the samples were analyzed for after storage quality.

Table 1. Storage treatments and units

Moisture Reduction Methods (A)	Storage temperatur (B)			
	3 °C (A1)		28 °C (A2)	
	Storage peiod (C)		Storage peiod (C)	
	2 months (C1)	4 months (C2)	2 months (C1)	4 months (C2)
Dehydration (A1)	4 bottles	4 bottles	4 bottles	4 bottles
Dehumidification (A2)	4 bottles	4 bottles	4 bottles	4 bottles

The moisture reduction stage was conducted using completely randomized design and reduction method (dehydration and dehumidification) was the treatment with four replications. The storage stage was conducted in 2x2x2 factorial experiment and the factors were moisture reduction method (dehydration/A1 and dehumidification/A2), storage temperature (3°C/B1 and 28°C/B2), and storage period (2 months/C1 and 4 months/C2). The quality consisted of moisture content, HMF, reducing sugar, diastase number, acidity (SNI 01-3545-1994), and total yeast (Fardiaz 1989). The data were analyzed by ANOVA and LSM at  $\alpha=0.05$  (SAS 1987) also by contrast test (SAS 1987) to determine changes in quality that occurred during storage.

## Results and Discussion

The effect of moisture reduction methods on the honey quality is presented in Table 2.

Table 2. The quality of raw and moisture reduced honey (inital storage honey)

Quality Variables	Raw Honey	Moisture Reduction Method		SNI 01-3545-2004
		Dehydration	Dehumidification	
Moisture (%)	21.60 <sup>a</sup>	17.60 <sup>b</sup>	17.50 <sup>b</sup>	max 22
Reducing sugar (%)	73.69 <sup>b</sup>	77.27 <sup>a</sup>	78.12 <sup>a</sup>	min 65
HMF (mg/kg honey)	37.28 <sup>c</sup>	46.40 <sup>a</sup>	41.22 <sup>b</sup>	max 50
Diastase number (DN)	4.69 <sup>b</sup>	3.93 <sup>c</sup>	5.74 <sup>a</sup>	min 3
Acidity (ml NaOH 1 N/kg honey)	39.22	38.22	36.10	max 50
Total yeast (cell/g honey)	7.6 <sup>a</sup>	2.7 <sup>b</sup>	4.2 <sup>b</sup>	-

Different superscript in the same row meant significantly different ( $P<0.05$ )

High HMF levels and acidity also low diastase number of raw honey could be the result of prolonged storage (7 months), but the low amount of yeast (7.6 cells/g of honey) indicated that the honey had been heated in advance to reduce the water content (21.60%). The moisture contents of the honey after being reduced by dehydration and dehumidification method (17%-18%) were still within the interval where honey was not susceptible to fermentation if the total yeast lower than 1000 cell/g honey. The decrease of

yeastin dehumidified honey due to the low moisture content (<18%), so that yeast cell became inactive (White 1992a). Repeated heating at 57 °C for an hour in the dehydration method in fact did not kill all yeast, but significantly increased HMF and damaged diastase enzyme. The formation of HMF was a result of decomposition of 6 C chained monosaccharides (glucose and fructose) which were reducing sugars (Cozmuta *et al.*, 2011), yet reducing sugar content in both dehydrated and dehumidified honey increased due to a decrease in water levels and increase the dry matter content of honey. The increase diastase number in dehumidified honey presumably because of a condition that supported transformation of inactive enzyme (zymogen) or the precursor of the enzyme into active enzyme (Winarno, 1985).

The changes on dehydrated and dehumidified honey quality after storage were summarized in Table 3. The moisture content was affected by interaction between the moisture reduction methods and storage temperature. Moisture content of dehumidified honey did not differ in the two storage temperatures, whereas dehydrated honey at 28°C was significantly higher than 3 °C. The higher moisture content was not caused bywater absorpsion from the surrounding since all honey samples were stored in sealed bottles, but due to yeast activity which was indicated by higher amount of yeast (6.7 vs 3.5 cells). Yeast digest carbohydrate and produce moisture and acid, resulting in increase moisture content and acidity of honey (Achmadi 1991). Contrast test showed that moisture content of dehydrated and dehumidified honey which were stored in 3°C until 4 months were not differ from the initial storage honey. Storage at a temperature of 3 °C is able to maintain dehydrated and dehumidified honey water levels remain low.

Table 3. The quality of dehydrated and dehumidified honey after stored in 3 dan 28 °C for 2 and 4 months

Quality Variables	Moisture Reduction Method(A)		Storage Temperature (B)		Storage Period (C)		Average
			3°C	28°C	2 months	4 months	
Moisture(%) (A*B) ; (C)							17.4 <sup>b</sup>
							17.7 <sup>a</sup>
							17.5 <sup>b</sup>
							17.5 <sup>b</sup>
							17.5 <sup>b</sup>
							17.6 <sup>a</sup>
HMF (mg/kg honey) (A) ; (B*C)							31.29 <sup>a</sup>
							25.59 <sup>b</sup>
							22.14 <sup>b</sup>
							24.26 <sup>b</sup>
							24.28 <sup>b</sup>
							43.06 <sup>a</sup>
Diastase number (DN) (A*B) ; (C)							4.16 <sup>a</sup>
							2.79 <sup>b</sup>
							4.17 <sup>a</sup>
							3.84 <sup>a</sup>
							3.45 <sup>b</sup>
							4.04 <sup>a</sup>
Reducing sugar (%) (A) ; (C)							77.70 <sup>b</sup>
							78.51 <sup>a</sup>
							76.07 <sup>b</sup>
							80.14 <sup>a</sup>
Total yeast (cells/g honey) (A) ; (B) ; (C)							3.2 <sup>b</sup>
							6.7 <sup>a</sup>
							3.5 <sup>b</sup>
							6.7 <sup>a</sup>
							3.4 <sup>b</sup>
							6.5 <sup>a</sup>
Acidity (mekNaOH 0.1N/kg honey) (B) ; (C)							38.10 <sup>b</sup>
							39.35 <sup>a</sup>
							38.35 <sup>b</sup>
							39.10 <sup>a</sup>

(A);(B); (C)=significantly different in the same factor (p<0.05)

(A\*B); (B\*C)= significantly different in the factor interaction (p<0.05)

The HMF content was affected by the moisture reduction methods, which was higher in dehydrated honey (31.29 vs 25.59) due to higher content in initial honey (Table 2). Interaction between the storage temperature and period also affected HMF content, the longer the honey stored the higher its HMF content, but the rate of increase was dependent on storage temperature. Honey stored in 28°C for 4 months contained the highest HMF (43.06 vs 22.14-24.28), because the temperature (even the room temperature), moisture (17.6-17.7), reducing sugar (80.14 vs 76.07), and acidity (around 39) which were higher in this treatment supported the formation of HMF (White, 1992b; Ribeiro *et al.*, 2012). Contrast test showed that dehydrated and dehumidified honey stored in 3°C until 4 months contained less HMF than the initial storage honey because low temperature inhibited HMF formation (Chai *et al.*, 1988) while the HMF that had been formed undergo further reactions and was changed into levulinic and formic acids (Achmadi, 1991).

Diastase number was affected by interaction between moisture reduction methods and storage temperature. Heating the honey in dehydration method resulted in instability of diastase enzyme activity which was indicated by significantly different diastase number between 3 °C (4.16) and 28 °C (2.79), *vice versa* with non-heating dehumidified honey which enzyme activity were stable in both temperature (Cozmuta *et al.*, 2011). Diastase number was also affected by storage period and significantly higher in 4 months storage (4.04 vs 3.45). Increase activity in this study could be triggered by the storage condition that supported the activation of enzyme's precursor in honey (Winarno, 1985), or there was recovery activity of the enzyme (Tosi *et al.*, 2008). Contrast test showed that the activity of diastase enzyme in dehydrated honey stored in 3 °C until 4 months was not significantly different from the initial honey. Conversely, the diastase number in dehumidified honey was significantly lower than the initial honey and this result was in line with Chai *et al.* (1988). Storage in 3 °C increased viscosity of dehumidified honey and blocked enzyme diffusion with substrate, thus lowered the activity of the diastase enzyme up to 4 months.

The reducing sugar content was affected by moisture reduction methods. Dehydrated honey contained lower reducing sugar than dehumidified honey (77.70 vs 78.51) due to lower diastase number (2.79) caused by heating in dehydration method (Cozmuta *et al.*, 2011). Diastase enzyme decomposed the complex carbohydrate into simpler carbohydrate and invertase enzyme hydrolyzed sucrose into glucose and fructose (Winarno, 1985). The low reducing sugar content in dehydrated honey was also caused by increased degradation of reducing sugar that has 6 carbon chain (glucose and fructose) into HMF hence the higher HMF content (31.29 vs 25.59). The reducing sugar content was also affected by storage period, which was higher in the 4 months stored honey (80.14 vs 76/07). Although HMF contents in the honey kept in 28°C or 4 months were high (43.06 vs 22.14-24.28), the diastase enzyme activity was also higher (4.04 vs 3.45). It seemed that the speed of reducing sugar formation exceeded its degradation into HMF, so the content of reducing sugar was increased. Contrast test showed that all honey stored in 3°C for 4 months contained higher reducing sugar than before the storage. Keeping dehydrated and dehumidified honey at 3°C until 4 months increased reducing sugar content through slowing down reducing sugar degradation into HMF.

Number of yeasts during storage was influenced by moisture reduction methods. Total yeast in dehydrated honey was lower than dehumidified honey though at the beginning it was not differ. The effectiveness of dehydration method in reducing the number of yeast was through the killing of most yeasts with high temperatures (57 °C), whereas dehumidification only inhibits the growth and development of yeast with low moisture content, so that number of yeast in dehumidified honey increased again during storage. The number of yeasts was also influenced by storage temperature, yeast growth at 28 °C were quick (6.5); on the contrary, it cannot thrive at a temperature below 11°C, hence the decrease in number at 3 °C (3.4) (Graham 1992). Storage time also affected the number of yeast, which was highest in four months, but the numbers was still below 1000 cells, so honey with 17% -18% moisture content was not susceptible to fermentation (White, 1992a). Contrast test showed that dehumidified honey stored in 3 °C for 4 months contained more yeast than it was at the beginning of storage due to increase reducing sugar which was yeast's food. This indicates that dehumidified honey should not be stored for a long time even in 3 °C.

The changes in the number of yeast during storage will affected acidity of honey due to yeast activity, hence the increase acidity at 28°C (39.35) and 4 months storage (39.10). This result was in accordance with the results of Chai *et al.* (1988) study. Contrast test showed that acidity of dehydrated honey stored in 3 °C did not differ from the initial storage honey, contrary to the dehumidified honey. The increase acidity in dehumidified honey was in line with the increase in the number of yeast which consumed reducing sugar and produced acetic acid (Achmadi, 1991).

## Conclusion

Dehydration method significantly decreased the number of yeast, but had negative effect due to increased HMF content and decreased activity of diastase enzyme. Dehydrated and dehumidified honey storage at a temperature of 3 for 4 months increased the content of reducing sugars and diastase enzyme activity without increasing HMF content, amount of yeast, and acidity of honey

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# Nitrite Residue and Sensory Characteristics of Dendeng With Addition of Strawberry (*Fragaria ananassa*) as Curing Agent

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## Abstract

Nitrite is used for curing as color stabilizer and antioxidant agent. Nitrite in meat product could affect human health by forming toxic compound (nitrosamine). Therefore, the use of nitrite should be substituted by natural antioxidant. Natural antioxidant such as ascorbic acid and phenol compound are contained in the fruits such as strawberry (*Fragaria ananassa*). The aim of this study is to evaluate the influence of strawberry as curing ingredient on nitrite residue and sensory characteristics of dendeng. The study was designed by using factorial treatment (2 x 4) with three replicates. The first factor is level of nitrite addition (0 ppm and 125 ppm) and the second factor is level of strawberry addition (0%, 10%, 20%, and 30%). The result showed that the addition of strawberry at 20% and 30% could reduce nitrite residue on product because it has lower value of nitrite residue than other treatment ( $P < 0,05$ ). Dendeng with addition of nitrite has better color; either based on hedonic or hedonic quality test ( $P < 0,05$ ). However, dendeng from all treatments has the same level for aroma, flavour, and texture. As conclusion the addition of strowberry up to 30% as curing ingredient on dendeng making could reduce nitrite residue, without affects the sensory charachteristic.

**Keywords:** dendeng (indonesian dried meat), nitrite residue, organoleptic characterictics, strawberry

## Introduction

Nitrate or nitrate salt is used for curing as color stabilizer and antioxidant agent on commercial dendeng making. Nitrate or nitrite could from  $\text{NO}_2$  and  $\text{NO}$  in meat system.  $\text{NO}_2$  react with water to form acid, and  $\text{NO}$  could react with myoglobin or amino acid compound. Reaction  $\text{NO}$  with myoglobin would form nitrosylhaemocromogen as red color pigment on cured meat (Honikel 2008). But, if  $\text{NO}$  react with secondary or tertiary amines it would form carcinogenic compound, nitrosamine (Suryati *et al.* 2014). The formation of nitrosamine could be inhibited by antioxidant.

Natural antioxidant such as ascorbic acid and phenol compound are contained in the fruits such as strawberry (Proteggente *et al.* 2002). Ascorbic acid as electron donor could react with reactive  $\text{NO}$  to form nitrite oxide, so it decreased reaction of reactive  $\text{NO}$  with amines. But, the use of strawberry on dendeng making is not popular because the effect on dendeng characteristics is unknown yet. The aim of this study was to evaluate theeffect of strawberry as curing ingredient on nitrite residue and sensory characteristics of dendeng.

## Materials and Methods

The study used meat obtained from knuckle of Brahman cross cattle. Dendeng was made by addition of  $\text{NaNO}_2$ , strawberry, salt, sugar, and the spices i.e. garlic, coriander, pepper, and galangal. The analyses of nitrite residue used acetic acid, 1-(N)-naphthylethylenediamine dihydrochloride and sulfanilamide. Dendeng was produced and prepared as sample according to the procedure described by Suryati *et al.* (2014) with difference on materials formulation (Table 1).

## Results and Discussion

Nitrite residue on dendeng ranged from 0.15 to 0.69mg  $\text{Kg}^{-1}$  of dry matter (Table 2). This amount was very low and categorized safe based onIndonesia's regulation stated that the maximum level of nitrite in product is 30 mg  $\text{Kg}^{-1}$  (BPOM, 2013). Nitrite level on dendeng was influenced by interaction of sodium nitrite and strawberry addition during curing process. The addition of 125 ppm nitrite did not influence nitrite residuelevel on dendeng added 10% and 20% strawberry, but it was higher on dendeng treated by

30% strawberry. The addition of 20% and 30% strawberry could reduce nitrite residue on dendeng ( $P < 0.05$ ) without sodium nitrite addition, but not on dendeng with sodium nitrite addition. Nitrite residue at dendeng control (without sodium nitrite) could come from meat, water and spices. The increasing of residue nitrite that in line with strawberry addition level might be influenced by the red color of extract from sample contained strawberry was read as nitrite residue by spectrofotometer analysis.

Table 1. Composition of ingredients on dendeng making (%)

Materials	Treatments							
	N0S1	N0S2	N0S3	N0S4	N1S1	N1S2	N1S3	N1S4
Meat	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0
Palm Sugar	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Sugar	20.0	20.0	20.0	20.0	20.0	20.0	20.0	20.0
Salt	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0
Garlic	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0
Coriander	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Pepper	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Galangal	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Nitrite (ppm)	0.0	0.0	0.0	0.0	125	125	125	125
Strawberry	0.0	10.0	20.0	30.0	0.0	10.0	20.0	30.0

Note: N0S1=without sodium nitrite and without strawberry; N0S2=without sodium nitrite with 10% strawberry; N0S3=without sodium nitrite with 20% strawberry; N0S4=without sodium nitrite with 30% strawberry; N1S1=with sodium nitrite and without strawberry; N1S2=with sodium nitrite and with 10% strawberry; N1S3=with sodium nitrite and with 20% strawberry; N1S4=with sodium nitrite and with 30% strawberry.

Table 2. Nitrite residue on dendeng with addition of sodium nitrite and strawberry in different level

Nitrite addition (mg Kg <sup>-1</sup> )	Strawberry addition (%)			
	0	10	20	30
	Nitrite residue (mg Kg <sup>-1</sup> of DM)			
0	0.99±0.25 <sup>a</sup>	0.70±0.21 <sup>ab</sup>	0.32±0.10 <sup>bc</sup>	0.15±0.12 <sup>c</sup>
125	0.35±0.07 <sup>bc</sup>	0.41±0.12 <sup>bc</sup>	0.38±0.11 <sup>bc</sup>	0.73±0.20 <sup>ab</sup>

Note: Means ± SD, means with different superscript letters are different ( $P < 0.05$ ), DM = dry matter

The color of dendeng with the addition of nitrite (N1) was preferred than dendeng without the addition of nitrite (N0) ( $P < 0.05$ ) (Figure 3). Nitrite had functions as a color stabilizer by forming a red color pigment, nitrosomyoglobin (Honikel, 2008). However other sensory properties of dendeng samples were not different.

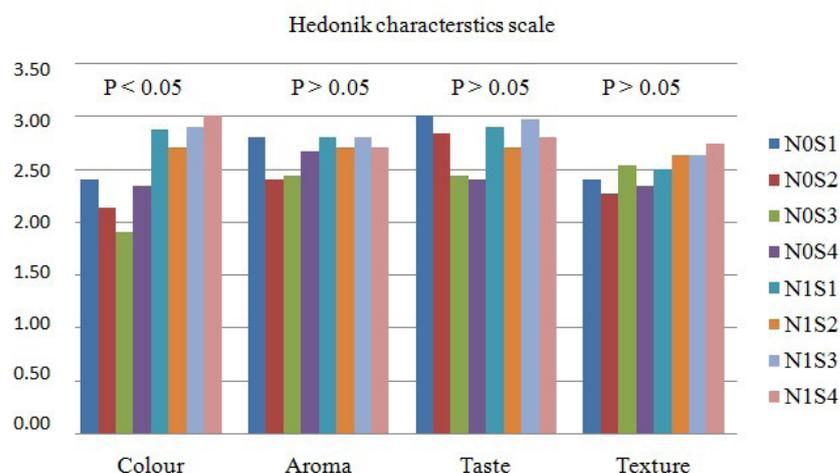


Figure 1. Hedonic characteristics of dendeng with addition of sodium nitrite and strawberry in different levels. Hedonic score: 1 = very dislike, 2 = dislike, 3 = like, 4 = very like.

Sensory characteristics based on hedonic quality of dendeng were represented at Figure 2. Dendeng added sodium nitrite in curing process had rather red and red colors. The red color is produced by the reaction of nitrite with myoglobin to form red pigment (Honikel 2008). The addition of strawberry in curing process of dendeng did not affect the red color. Anthocyanin or red pigment in strawberry had a low stability. The red color of anthocyanin appear at pH 1-3 and unstable in heat during processing (Markakis, 1984). Sensory properties such as aroma, taste, and texture of all treatments were not different. Aroma and taste of dendeng had rather sour to not sour. The rather sour aroma and taste of dendeng treated by strawberry addition comes from organic acids and vitamin C contained in strawberry (Koyuncu dan Dilmacunal, 2010). Texture of dendeng was rather tough (not tough), it was caused by rehydration process during soaking in water before frying. During rehydration water was absorbed so that the water content and water activity increased.

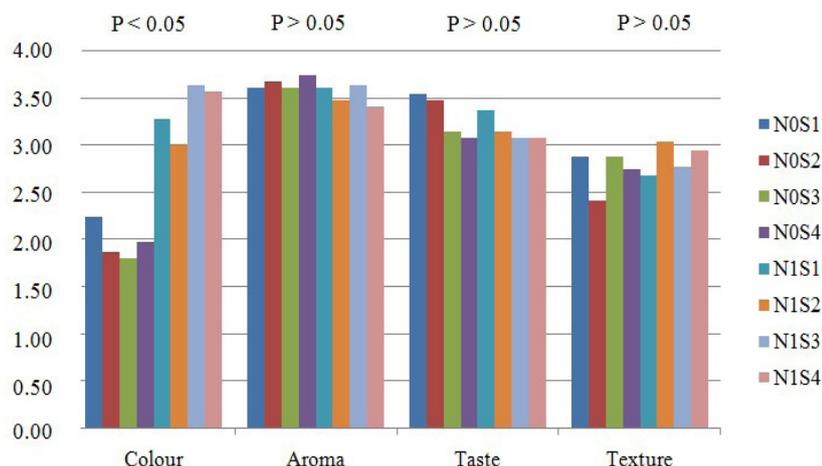


Figure 2. Hedonic quality of dendeng with addition of nitrite and strawberry in different level. Intensity scale of color (1= black, 2= rather black, 3= rather red, 4= red), aroma (1= very sour, 2= sour, 3= rather sour, 4= not sour), taste (1= very sour, 2= sour, 3= rather sour, 4= not sour), texture (1= very tough, 2= tough, 3= rather tough, 4= not tough).

## Conclusion

The addition of strawberry from 20% to 30% could reduce nitrite residue on dendeng that comes from environment, but not for dendeng added by sodium nitrite. Strawberry addition resulted in fresh rather sour aroma and taste on dendeng. The use of strawberry did not increase red color intensity of dendeng as like the use of sodium nitrite.

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# Biodiversity Based on Flavor and Amino Acid Profile of Indonesia Local Chickens

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## Abstract

Indonesia has rich of flora and fauna biodiversities. One of the fauna that has the potential to be developed is a local chicken. Local chicken also called native has advantages over the chicken, including disease resistance, typical and preferred meat flavor, and has several health benefits. In terms of nutritional, local chicken meat contains protein which is not much different from the protein in chicken meat and fat content is lower. The taste and flavor was associated with fatty acid composition of meat. However, data on the local Indonesian chicken excellence as a whole has not been published in full. As a result, the advantage of Indonesian local chicken is not known well. It is very interesting if known diversity of Indonesian natural resources, especially fatty acid and amino acid profile on various types of typical Indonesian local chicken. This research was conducted with a purposive sampling method on chicken farms in Indonesia regardless of feed given. This study was conducted to assess the biodiversity of local chicken excellence in several regions in Indonesia. Chicken samples were derived from four different island in Indonesia: Banyuwangi (East Java), Bogor (West Java), Jambi (Sumatra) and Makassar (South Sulawesi). Principal Component analysis (PCA) was used for describing fatty acid profile of local chicken. The results obtained showed that the chicken from Bogor (West Java) has the best quality hedonic quality ( $p > 0.05$ ) compared with the other chickens. Chicken from Jambi has highest fatty acid content (70.46 %) and best mayor amino acid profile. Principal Component Analysis of fatty acid profile of breast Indonesian local chicken showed that there was principal variance of fatty acid as 71.1%, and can be analysed with equation:  $Y_1 = 0.476$  Alcohol (1-Hexanol) + 0.153 Keton (3-Octanone, 2-methyl) + 0.415 Furan + 0.474 Hydrocarbons)Tridece + 0.484 Hydrocarbone (Tetra) + 0.344 Aldehyde (Nonanal). The correlation alcohol was 0.98. There are mayor amino acid profile of breast Indonesian local chickens such as aspartic acid, glutamic acid, serine, hidtidie, glycine, threonine, arginine, alanine, tyrosin, methionine, valine, phenylalanine, I-leucine, leucine and lycine.

## Introduction

Indonesia has rich of flora and fauna biodiversities. One of the fauna that has potential to be developed is a local chicken. Local chicken also called native chicken with has advantages over the chicken, including disease resistance, typical and preferred meat flavor, and has several health benefits. According to statistical data of Ministry of Agriculture Republic of Indonesia, chicken population in Indonesia in 2010 reached more than 277.4 million. This amount can contribute 40% of total production of poultry meat and 13.8% of the insufficient demand for animal protein.

In terms of nutrients, chicken meat contains protein which is not much differ from the protein in chicken meat and lower fat content. The fat content of local chicken meat is lower than broilers. Local chicken is more attractive to the public than the chicken because of taste. Taste and flavor associated with the fatty acid composition of the meat. However, data on the advantages of local chickens Indonesia as a whole has not been well published yet. As a result, the advantage of Indonesian local chicken is not known well. It is very interesting if known diversity of Indonesian natural resources, especially fatty acid and amino acid profile on various types of typical Indonesian local chicken.

This study was conducted to assess the biodiversity advantage of local chicken of Indonesia (4 types of chicken from Banyuwangi, East Java, Jambi-Sumatra, Ciampea- West Java, and Makassar - South Sulawesi) in terms of flavor and amino acid composition.

## Materials and methods

This study was conducted with a purposive sampling method on chicken farms in Indonesia regardless of feed given. A total of six local female chickens adult age (6-7 months), respectively sampled. Local

chickens were sampled from extensive production system. Chicken meat samples have been collected and analyzed. Flavor analysis was conducted by Gas Chromatography, while amino acid composition was determined by using high-performance liquid chromatography (HPLC). The procedures were according to AOAC (2005). Data was analyzed statistically by ANOVA and correlation analysis (Steel & Torrie 1995). Principal Component analysis (PCA) was used for describing fatty acid profile of local chicken.

## Result and Discussion

Flavor analysis using GC-MS yielded some of flavor component as described in Table 1.

Table 1. Flavor profile of Indonesia local chicken meats from different areas.

Flavor component	Banyuwangi	Bogor	Makassar	Jambi
Alcohol	17.04±2.85 a	22.34±7.77 a	3.22±2.83 b	1.79±0.06 b
Aldehyde	11.39±4.38 a	16.05±7.05 a	2.86±0.43 b	0.84±0.58 c
Keton	7.77±3.39 a	7.86±2.22 a	3.49±0.50 b	0.83±0.08 c
Carboxylic acid	0.03±0.01	0.11±0.01	0.03±0.06	0.11±0.02
Furan	0.72±0.42 a	0.90±0.42 a	0.14±0.01 b	0±0 c
Hydrocarbon	9.88±5.14 b	16.03±5.14 a	5.28±2.38 c	3.15±2.99 c
Aromatic hydrocarbon	0.14±0.02 a	0.06±0.02 b	0.15±0.05a	0±0 c
Nitrogen contain	0±0 c	0.11±0.08 a	0.06±0.1 b	0±0 c

Note: Means ± SD, means with different superscript letters are different (P < 0.05)

Results of the flavor analysis in Table 1 shows significant differences (P < 0.05) on some the value of flavor components in chicken meat that comes from several different areas in Indonesia, such as alcohol, aldehyde, keton, furan, hydrocarbon, aromatic hydrocarbon and nitrogen contain. Flavor component of local chicken from Bogor has highest flavor component percentage, while local chicken from Jambi has lowest flavor component. The result showed that environment such as climate, and local feed contribute the varieties of flavor component of local chicken in Indonesia (Fletcher 1999).

Principal component analysis of flavor profile of breast Indonesian local chicken showed that there was principal variance of flavor component as 71.1%, and can be analysed with equation :  $Y_1 = 0.476 \text{ Alcohol (1-Hexanol)} + 0.153 \text{ Keton (3-Octanone, 2-methyl)} + 0.415 \text{ Furan} + 0.474 \text{ Hydrocarbons)Tridece} + 0.484 \text{ Hydrocarbone (Tetra)} + 0.344 \text{ Aldehyde (Nonanal)}$ . The correlation alcohol was 0.98.

Results of amino acid composition in Table 2 shows no finding significant differences (P > 0.05) of chicken meat obtained from several different areas in Indonesia. That means, local chicken has no variety on amino acid composition effect to protein content of meat. There are mayor amino acid profile of breast Indonesian local chickens such as aspartic acid, glutamic acid, serine, hidtidie, glycine, threonine, arginine, alanine, tyrosin, methionine, valine, phenylalanine, I-leucine, leucine and lycine.

Table 2. Amino acid composition of local Chicken meat from different areas in Indonesia

Amino acid	Banyuwangi	Bogor	Makassar	Jambi
Aspartic Acid	2.24±0.26	1.91±0.32	2.09±0.23	2.15±0.39
Glutamic Acid	4.66±1.41	3.34±0.54	3.67±0.50	3.79±0.64
Serine	1.02±0.12	0.88±0.15	0.98±0.08	0.95±0.17
Histidine	0.71±0.11	0.72±0.14	0.76±0.07	0.70±0.18
Glycine	1.14±0.28	0.95±0.23	0.94±0.12	1.13±0.22
Threonine	1.08±0.13	0.92±0.15	1.04±0.11	1.02±0.19
Arginine	1.64±0.20	1.33±0.23	1.51±0.16	1.53±0.27
Alanine	1.47±0.17	1.25±0.23	1.34±0.11	1.42±0.24
Tyrosin	0.86±0.11	0.72±0.12	0.84±0.10	0.79±0.14
Methionine	0.62±0.10	0.52±0.07	0.56±0.09	0.55±0.04
Valine	1.16±0.13	0.98±0.17	1.11±0.10	1.07±0.22
Phenylalanine	1.02±0.12	0.84±0.14	0.96±0.09	0.95±0.19
I-Leucine	1.15±0.14	0.94±0.16	1.09±0.11	1.05±0.21
Lecine	2.00±0.24	1.66±0.28	1.88±0.17	1.86±0.36
Lycine	2.10±0.32	1.66±0.31	1.72±0.41	1.90±0.43

Note: Means ± SD

## Conclusion

Local chickens from different areas in Indonesia has diversity on flavor component of breast meats, but no variety on amino acid profile. Local chicken from Bogor-West Java has highest flavor component, while local chicken from Jambi is lowest. PCA of flavor profile of breast Indonesian local chicken showed that there was principal variance of flavor component as 71.1%.

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# Moisture, pH Value and Physical Quality Stability of Dendeng During Storage at Different Temperature

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## Abstract

Dendeng, Indonesian dried meat, could decrease its quality during storage. This study evaluates moisture, pH value and physical stability of dendeng during storage (for 12 days) at different temperature (29°C, 37°C or 45°C). Dendeng made from fresh beef, sliced 5 mm, cured with mixed spices, and dried by oven (60°C for 3 hr then continued at 70°C for 6 hr) were packed in sealed plastic, and then it was kept in oven at 29°C, 37°C or 45°C for 12 days. The variables which covered moisture, pH value, water activity, and sensory characteristic (color, rancid flavor and the existence of mold) were evaluated every 3 days. The results showed that moisture, pH value, water activity, and sensory characteristics of dendeng kept at each temperature were not different during storage for 12 days. That variables also were not different among storage at 29°C, 37°C or 45°C for 12 days. The range of moisture, pH value, and water activity were 19.34% to 23.54%, 4.84 to 5.54, and 0.629 to 0.734, respectively. For all treatments, the color was still in normal range (reddish brown to brown), the rancid flavor was not detected significantly, and the mold was not appeared. It could be concluded that dendeng quality was stable at 29°C, 37°C or 45°C for 12 days.

**Keywords:** dendeng (Indonesian dried meat), stability, storage, temperature

## Introduction

Meat processing to be dendeng could increase shelflife of product through the inhibition of oxidation reaction (Suryatiet *al.* 2014) and deterioration (Bintoroet *al.*, 1987) in product. Nevertheless, the quality of dendeng could decrease during storage, either physically or chemically, till it couldn't be consumed indicated by strong rancid flavor or existence of mold. Decrease of quality is held caused by increasing of moisture, pH value and water activity ( $a_w$ ) resulting oxidation reaction, non enzymatic browning and microbe growth more intense. That process impact dark color, offodor or rancid flavor, and deterioration ondendeng. This study was held to evaluate stability of beef dendeng quality during storage at different temperature.

## Materials and Methods

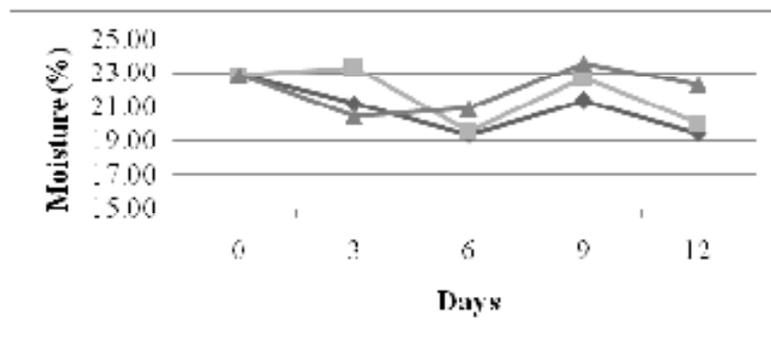
Dendeng was made from beef with ingredient composition and method according to Suryatiet *al.* (2014). Total drying duration was modified to be 9 h. Dendeng was packed by using LDPE plastic and storage at 28°C, 37°C and 45°C for 12 d. Evaluation of moisture, pH value,  $a_w$ , and sensory characteristics were held at 0, 3, 6, 9 and 12 d. Moisture was analyzed based on oven method following AOAC (2005). pH value was determined using pH meter Hanna HI 99163 (Romania, Europe), and  $a_w$  was measured by  $a_w$  meter type ms1 Set-aw (Novasina, Switzerland). Sensory characteristics evaluated covered intensity of color, rancidity flavor and existence of mold.

## Results and Discussion

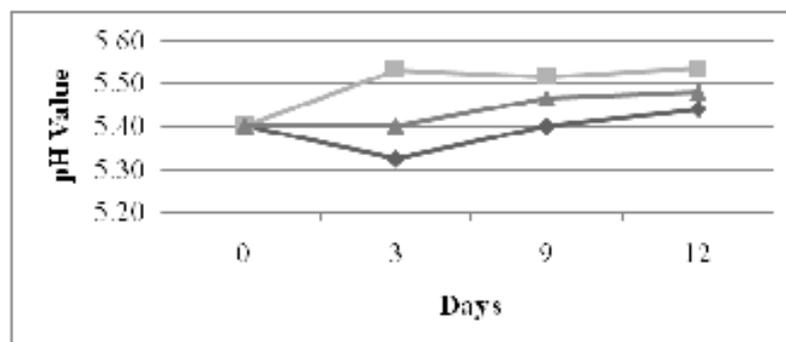
The range of moisture during 12 d storage was 19.34% to 23.54%. Moisture of dendeng had fluctuation during storage at all temperatures (Figure 1). The storage at 28°C and 37°C had similar pattern, the moisture at 0 to 6 d of storage decreased up to about 19%, and then increased till to 9 d and decreased again until 12 d storage. The storage at 45°C had rather different pattern from at 28°C and 37°C storage, decreasing of moisture took place at 0 d up to 6 d, and then rising again up to 12 d similar to 28°C and 37°C storage. Generally the moisture of dendeng during 12 d storage was still in normal range (13.52% - 33.09%) as reported Suryatiet *al.* (2012).

Increasing of pH value generally was taken place during storage, except the storage at 28°C that it had lowering of pH value till to 3 d storage and rising again till to 12 d storage (Figure 2). The pH value of dendeng during storage was ranged 5.33-5.54. This range was still in normal according to pH value of commercial dendeng (5.13-5.66) (Suryati *et al.*, 2012).

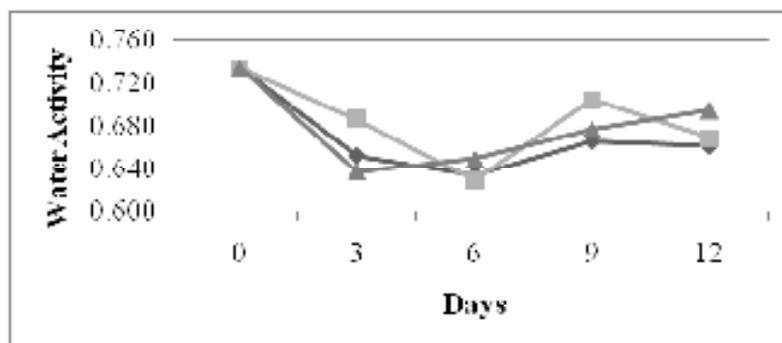
Decreasing of  $a_w$  generally was occurred begin from 0 d to 12 d storage with a little fluctuation (Figure 3). Water activity of dendeng for all temperature storage decreased to 3 d storage, and continued up to 6 d for 28°C and 37°C storage, but the storage at 45°C increased again up to 12 d storage with the value was still lower than initial  $a_w$ . Water activity of dendeng at 28°C and 37°C storage increased till 9 d and then decreased till 12 d storage. Water activity of dendeng for all temperature storages were 0.629-0.734. This range was still normal (Bintoro *et al.*, 1987; Suryati *et al.*, 2013).



**Figure 1. Moisture of dendeng during storage for 12 d at 28°C (♦); 37°C (■); 45°C (▲)**



**Figure 2. pH value of dendeng during storage for 12 d at 28°C (♦); 37°C (■); 45°C (▲)**



**Figure 3. Water activity of dendeng during storage for 12 d at 28°C (♦); 37°C (■); 45°C (▲)**

The color intensity of dendeng up to 6 d storage for all temperatures increased from slightly darkness brown (score 3) to slightly redness brown (score 4), and then to be slightly darkness brown again at 12 d for 28°C and 45°C storage, but dendeng stored at 37°C had darkness brown color (score 2) at 12 d storage (Figure 4). It is generally that the color of dendeng for 12 d storage at 28°C, 37°C and 45°C ranged from darkness brown to slightly redness brown. This color range for dendeng without curing using nitrat/nitrit salt was normal as reported by Suryati *et al.* (2013).

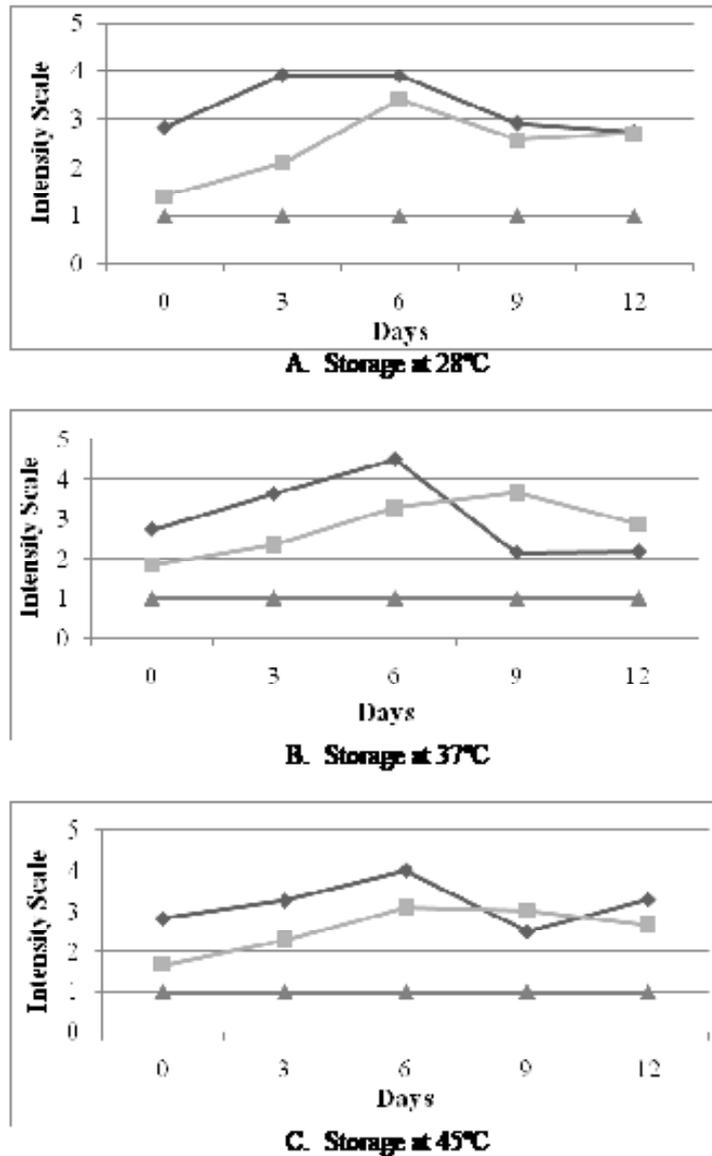


Figure 4. Sensory characteristics of dendeng during storage for 12 d. A. Storage at 28oC; B. Storage at 37oC; C. Storage at 45oC. Colour intensity(-◆-): 1=extremely dark; 2=darkness brown; 3=slightly darkness brown; 4=slightly redness brown; 5=redness brown; 6=brownness red; 7=slightly brownness red; 8=red. Rancidity flavor intensity (-■-):1=extremely weak; 2=very weak; 3= weak; 4=slightly weak; 5=slightly strong; 6=strong; 7=very strong; 8=extremely strong.Mold existence (-▲-): 0=no exist; 1= exist.

Rancid flavor intensity of dendeng for 12 d storage at at 28°C, 37°C and 45°C was ranged from extremely weak to slightly weak (Figure 4). This indicated that the rancid flavor of dendeng was not smelt significantly by panelist. The growth of mold in dendeng up to 12 d for all temperatures storage was not visible (Figure 4). Based on rancid flavor intensity and the existence of mold indicated that quality of dendeng for 12 d storage at 18 °C, 37°C and 45 °C were fine.

## Conclusion

Quality of beef dendeng in this study was stable at 28°C, 37°C or 45°C for 12 days.

## Acknowledgement

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# Milk Production of Sahiwal x Holstein Crossbreed in Two Different System on Local Farm Kudat, Sabah-Malaysia

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## Abstract

In Sabah-Malaysia there are two dairy farming systems, i.e. intensive and semi-intensive system. In intensive system, the animals kept on colony stable, meanwhile in semi-intensive system, the animals are reared on integrated oil palm plantation. The objective of this research was to compare the two different farming systems on nutrient intake and performance of dairy cattle in local farm Kudat, Sabah-Malaysia. In this research, 90 heads of Sahiwal x Holstein Crossbreed dairy cows from 4 farmers were observed for two month in September 2012. The parameters observed were feed consumption, milk production, body weight and body condition score (BCS). The data were analyzed using T-test. The results showed that daily dry matter intake on intensive system were higher than on semi-intensive system (15.87 and 10.16 kg head<sup>-1</sup> day<sup>-1</sup> respectively). The BCS of lactation cows of both systems was no significantly different (3.08 vs 2.95). However, the average milk production on intensive and semi-intensive system showed significant differences, were 19.7 and 18.1 head<sup>-1</sup> day<sup>-1</sup> respectively. Cost of feed (MYR head<sup>-1</sup> day<sup>-1</sup>) in two different farming was significantly different (37.20 MYR on intensive system and 38.70 MYR on semi-intensive system). Base on income over feed cost (IOFC) in both systems, it is concluded that intensive system was more profitable than the semi-intensive system.

Keywords: dairy cattle, dairy farming system, milk production, Sahiwal x Holstein crossbreed.

## Introduction

In Sabah-Malaysia, the dairy cattle development was started in Keningau and Tawau from 1981-1985. According to Salleh (1989), this project involved small land holders with the intention of forming the dairy industry and thereby increasing urban resident's income in conjunctions with the new economic foundation. Dairy industry in Sabah is growing forward to increase milk production to reached 10 million liter per year. Now, Sabah is a major producer of fresh milk for public school milk program.

Main breed of dairy cattle in Sabah is SxF crossbreed, which crossing from local Sahiwal dairy cattle with Australian and New Zealand Frisian Holstein (Sivarajasingam *et al.* 1983). SxF is best performance regarding to milk composition and quality (Talukder *et al.* 2013). In Sabah this crossbreed is called Sabah Sahiwal Friesian (SSF).

Dairy cattle farming management on local farm Kudat are intensive and semi-intensive system. In the intensive system, dairy cows kept in colony stable all day and fed according to nutrient requirement. While in the semi-intensive system, cows grazing in the oil palm plantation or rubber plantation during the day and in the evening brought to the stable.

A problem in dairy cattle production in Sabah is limited land for grass production. Meanwhile, the oil palm and rubber plantations have the potential to supply forage. Forage growing under the palm trees can be used as animal feed by cutting or used as a grazing area. However, the quality of forage under oil palm plantations is poor. Differences in production systems has an impact on the adequacy of nutrients supply to the dairy cows.

The objective of the study was to study the nutrient intake and performance of dairy cattle on two different production systems on local farm Kudat, Sabah-Malaysia.

## Materials & Methods

### Location

The study was carried out in Kudat, Sabah, Malaysia to evaluate the differences milk production and performance on intensive and semi-intensive dairy production systems.

## Animals

In this study, 90 heads of crossbreed of Sahiwal x Friesian (SxF) dairy cattle from both intensive and semi-intensive farms were observed. The animals were belongs to 4 farmers from different locations.

## Feeding Management

On intensive dairy production system, the animals were kept in colony stable and fed with cut napier grass (*Pennisetum purpureum*), palm kernel cake (PKC) as concentrate source and mineral block as a feed supplement. Meanwhile, on semi-intensive system, the animals were kept on grazing area under oil palm plantation for 6-9 hours in a day and in the evening the animals were kept in stable and fed some napier grass and PKC. In both group farms, the dairy cows were milked using milking machine twice a day.

## Methodology

This study was conducted by interview and observation directly atlocal dairy farmers. The variables measured were body weight of cows, body condition scoring (BCS), feed intake (grass and concentrate), milk production and income over feed cost.

Data were analyzed using T-test to compare the variables between two different farming systems using the Statistical Package for the Social Sciences (SPSS).

## Results & Discussion

Dry matter (DM) and nutrient intake of dairy cows on intensive and semi-intensive farming systems are shown in Table 1. The result showed that DM forage intake on intensive system was significantly higher than on semi-intensive system (7.31 vs 2.81 kg DM head<sup>-1</sup> day<sup>-1</sup>). PKC intake of dairy cattle on intensive farming system was also higher than on semi-intensive farming system (9.24 vs 7.93 kg DM head<sup>-1</sup> day<sup>-1</sup>). The use of PKC on both farming systems was more than 50% in the ration. According to Zahari & Farid (2011) in dairy cattle rations, PKC can be used as a source of energy and protein at the inclusion level of 30-50%.

The average of DMI of dairy cows in intensive systems was significantly higher than in the semi-intensive system (15.87 and 10.16 kg<sup>-1</sup> head<sup>-1</sup> day<sup>-1</sup> respectively). On intensive farming system percentage of DMI of body weight (BW) was sufficient (3.05% of BW), while on semi-intensive farming system was very low (2.03% of BW). This lower DMI in semi-intensive farming caused by lower total nutrient digestible (TDN) intake (6.14 kg<sup>-1</sup> head<sup>-1</sup> day<sup>-1</sup>). According to NRC (2000), daily DMI and percentage of DMI of BW for the dairy cattle is about 3.0% of BW.

Table 1. Dry matter and nutrient consumption of dairy cows on intensive and onsemi-intensive farms in Sabah-Malaysia

Variables	Intensive System	Semi-intensive System	P-value
Forage intake (kg DM <sup>-1</sup> head <sup>-1</sup> day <sup>-1</sup> )	7.31±0.95	2.81±1.04	< 0.01
PKC intake (kg DM <sup>-1</sup> head <sup>-1</sup> day <sup>-1</sup> )	9.24±1.49	7.93±1.41	< 0.01
Total DM intake (kg DM <sup>-1</sup> head <sup>-1</sup> day <sup>-1</sup> )	15.87	10.16	-
CP intake (g <sup>-1</sup> head <sup>-1</sup> day <sup>-1</sup> )	2.21	1.15	-
CF intake (g <sup>-1</sup> head <sup>-1</sup> day <sup>-1</sup> )	3.12	1.33	-
TDN intake (kg <sup>-1</sup> head <sup>-1</sup> day <sup>-1</sup> )	10.35	6.14	-
Feed intake / body weight (%)	3.05	2.03	-

Table 2. Dairy cow performances and IOFC on intensive and onsemi-intensive farms in Sabah-Malaysia

Variables	Intensive System	Semi-intensive System	P-value
Body weight (kg)	520±100	499±32	-
Body condition score (BCS)	3.1±0.28	2.9±0.21	-
Milk production (liter head <sup>-1</sup> day <sup>-1</sup> )	19.7±1.1	18.1±0.5	< 0.01
Total Feed Cost (MYR head <sup>-1</sup> day <sup>-1</sup> )	37.20	38.70	-
Income Over Feed Cost (MYR head <sup>-1</sup> day <sup>-1</sup> )	12.10	8.02	-

The dairy cattle performance and IOFC is shown in Table 2. Body weight (BW) and body condition score (BCS) of dairy cattle on intensive system were relatively higher. This indicated that the animals kept on intensive system had better body condition than animals kept on semi-intensive system. Generally, cattle on pasture have lower BCS than cattle managed under a non-grazing environment (Lawrence, 2003).

The average of milk production of SxF dairy cattle on both farming systems was not much different, 19.71 kg<sup>-1</sup> head<sup>-1</sup> day<sup>-1</sup> on intensive farming system and 18.1 kg<sup>-1</sup> head<sup>-1</sup> day<sup>-1</sup> on semi-intensive farming system. This milk production was higher than reported by Boniface *et al.* (2007) that the average dairy cattle in Sabah was only 8.64 liters head<sup>-1</sup> day<sup>-1</sup>.

Although DMI of dairy cows on a semi-intensive system was very much different with on semi-intensive farming system, however milk production on both systems was not much different. This is probably the dairy cattle that grazing on semi-intensive farming system was still get sufficient forage.

Total Feed Cost in semi-intensive farming system was slight higher than in intensive farming system (38.70 and 38.70 MYR head<sup>-1</sup> day<sup>-1</sup>, respectively), This is because of cost of pasture management in semi-intensive system was high. Finally, income over feed cost (IOFC) on intensive system was higher than on semi intensive farming system.

## Conclusion

Dry matter and nutrient intake of dairy cattle on intensive farming system in Sabah were higher than on semi-intensive system, and then result more milk production. According to income over feed cost, dairy cattle on intensive farming systems in Sabah was more profitable than semi-intensive system.

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# Physical Meat Quality of Kacang Goat and Garut Sheep Fed Sorghum Based Concentrate

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## Abstract

Physical characteristics of goat and sheep meat (lamb) become an important criteria for consumers in ruminants meat products purchasing. The quality of meat depend on meat colours, tenderness, water holding capacity and cooking loss, it is also influenced by the type of feed and feeding factors. This study designed to evaluate local goat meat and lamb quality based on physical characteristics. Goats and sheeps were given sorghum based concentrate which contain high protein and carbohydrate. Six local Kacang goats and five Garut sheeps were fed concentrate with 20% sorghum and raised for 100 days. Measured physical characteristics were pH, water holding capacity, tenderness, cooking loss and meat colours. Data analyzed using ANOVA and t-test. There were significant differences ( $p < 0.05$ ) in pH  $6.03 \pm 0.10^a$  and  $5.794 \pm 0.085^b$ , tenderness  $2.733 \pm 0.33^a$  and  $1.860 \pm 0.23^b$  respectively between goat meat and lamb. Other parameters of goat meat and sheep meat were similar. Sorghum in goat and sheep feeding resulting good meat quality of both goat and sheep.

Keywords : goat meat , physical characteristics, sheep meat, sorghum

## Introduction

One effort to meet people's needs for animal protein is by breeding and raising different kinds of livestock, including goats and sheep. Indonesian local livestock such as Kacang goats and Garut sheep has a huge potential to be developed since these animals have several advantages compared with other kinds of livestock, for example, they breed rapidly and adjust to the environment easily.

In general, nutritionists consider meat an important part of a well-balanced diet because it provides protein, vitamins, minerals and fat necessary which are necessary for good health and growth. Meat protein contains essential amino acids needed to build and maintain body tissue. Meat is rich in iron, which is needed to build and maintain red blood cells and muscle growth. Red meat (meat of cattle, pigs, goats, sheep, etc.) is an excellent source of the vitamin B complex groups (B1, B2, B6, and B12).

All kinds of meat sold in traditional markets and supermarkets have to meet government health standard. That is why meat is graded according to its quality. Higher grades of meat are tenderer, juicy, and flavorful than lower grades. Grading is based on such quality factors as genetic, species, breed, sex, age, and diet (before slaughter), and withering method and meat pH (after slaughter) (Lambe, 2008).

The physical quality of meat, which consists of color, tenderness, water holding capacity, and cooking loss, is a consumer reference in buying meat. Water holding capacity greatly affects the appearance of the meat before cooking, its properties during cooking, and its juiciness when chewed (Lawrie, 2003). The quality of meat will increase as the water binding ability of the meat increases, lowering the cooking loss and lessening the loss of nutrients. The quality of the meat which is the end result of the fattening of local goats and sheep cannot be separated from the quality of the feed given. Feed is one of the factors that determine the quality of the meat. Feed management and nutrient content are contributing factors to obtain the good results of livestock production.

Sorghum (*Sorghum bicolor* L.) is one type of cereal crops that has great potential to be developed in Indonesia in view of its wide area of adaptability. Sorghum plants are tolerant to drought and water puddle, relatively resistant to pests / diseases, and can produce on marginal land. Sorghum is a carbohydrate source with a metabolic energy content of  $3,212 \text{ kcal kg}^{-1}$  (NRC 1994). In addition, sorghum has high protein content (12.99%) and low fat (2.34%) compared to corn. Sorghum grains are potential to be used as feed concentrates. Therefore, this study was conducted to determine the quality of meat produced by local goats and sheep which were reared intensively fed sorghum-based concentrates. The objective of this study was to determine and evaluate the meat quality values of Kacang goat and Garut sheep which were fed sorghum-based concentrate in an intensive rearing.

## Materials and Methods

Five male Garut sheep and six male Kacang goats less than one year of age were intensively reared for 100 days in an individual stall. The feed was in the form of sorghum-based concentrate (20% sorghum grain) mixed molasses and forage of *Brachiaria humidicola* (RBH) grass. The feeding consisted of 60% concentrate and 40% forage. Concentrate feed was given in the morning as much as 500 g / head, mixed with molasses 250 g / head. Forage was given in the afternoon 1kg / head and in the late afternoon 1 kg/head. A carcass part called *M. Longissimus dorsi* would be analyzed to test the physical quality, the observed variables such as pH test of meat, DMA, cooking loss, meat tenderness and color. The value of meat pH was measured by using a pH meter, and the water holding power was determined by using Hamm Formula (1972). The meat tenderness was measured objectively by using a tool called Warner-Bratzler shear. The meat color was tested objectively using Chromameter with hunter notation, that is, L \*, a \* b \*. The data obtained were analyzed using t-test.

## Results and Discussion

The meat quality of Kacang goat and Garut sheep intensively fed sorghum-based feed for 100 days based on their physical properties included pH value, DMA, tenderness, cooking loss and meat color, as can be seen in Table 1.

Table 1. Mean of physical quality of the meat of Kacang goat and Garut sheep fed sorghum-based feed

Type of Livestock	pH	DMA (%)	Cooking Loss(%)	Tenderness (kg/cm <sup>2</sup> )	Meat Color		
					L*	a*	b*
Kacang goat	6.03 <sup>a</sup> ±0.10	52.12±10.54	39.160±3.54	2.733 <sup>a</sup> ±0.33	45.77	13.96	5.243
Garut sheep	5.79 <sup>b</sup> ±0.08	44.80± 3.78	37.744±4.79	1.860 <sup>b</sup> ±0.23	43.78	12.23	5.164

<sup>a,b</sup>Different letters in the same column show a significant difference (P<0.05)

### pH Value

There was a significant difference (P> 0.05) between the pH value of Kacang goat meat and Garut sheep meat fed sorghum-based feed. The mean of pH value of each animal was 6.03 ± 0.10<sup>a</sup> and 5794 ± 0.085<sup>b</sup>, respectively. The measurement of pH value was done 24 hours after slaughter to determine the final pH which was achieved when the glycogen content of the meat was really exhausted. The decrease in meat pH during slaughter is influenced by lactic acid. The process of change from muscle into meat requires glycogen as an energy source and will produce lactic acid. This process causes glycogen to convert to lactic acid until the pH reaches a point when the breaker enzymes become inactive. The breaker enzymes (glycolytic) on specific mammalian meat will stop at a pH of 5.4 - 5.5 and in this condition glycogen cannot be found anymore in meat (Lawrie, 2003). The more lactic acid is available, the greater the decline of the meat pH of during slaughter and the lower the final meat pH. That the final pH value of Kacang goat meat will be higher than that of Garut sheep meat is due to the different glycogen content between the cattle. The glycogen content in Garut sheep meat is higher than in Kacang goat meat.

The goat or sheep that is quiet during slaughter has enough glycogen reserves for rigor mortis process, while the stressed one is likely to produce higher ultimate meat pH because the muscle of glycogen reserves gets exhausted quickly. The treatment of cattle before slaughter greatly affects them in order not to experience high stress during slaughter. To lower the stress level in cattle, before slaughter is conducted, the animal should be avoided doing a lot of activities. Good cattle handling before the slaughtering process will also contribute to the calmness of the cattle during slaughter.

### Water Holding Capacity (DMA)

The result showed that in the meat section of *longissimus dorsi* there was no significant difference in the DMA value of Kacang goat meat and Garut sheep meat. However, the DMA value of each animal was relatively high, that is, 52.120± 10.54% and 44.806 ± 3.78% respectively, so that the quality of both meat based on the value of their DMA was in the category of good. One of the factors that cause the high DMA value was that the cattle were relatively young, less than one year. The high water binding power of meat protein increases the tenderness and juiciness of the meat and decreases its cooking loss, lowering the loss of nutrients. DMA in meat is influenced by differences in muscle, species, breed, age, muscle function, sex,

intramuscular fat and storage temperatures (Soeparno, 1994). The percentage of the water that comes out of the meat can be used as an indicator to determine the value of DMA. The smaller the percentage of water that comes out of the meat, the higher the DMA value. The decrease in the DMA of beef and mutton is due to the formation of aktomiosin and the depletion of ATP at the time of rigor. A third of the DMA reduction in meat is caused by a decrease in pH. DMA is closely related to the cooking loss of the meat.

### **Cooking Loss**

Cooking loss during the cooking process is one indicator of the nutritional value of the meat. The higher the cooking loss of the meat, the more nutrients will lose. The analysis result of cooking loss based on t test showed that in the meat part of *Longissimus dorsi* there was no significant difference in the cooking loss values of the meat of Kacang goat and Garut sheep,  $39.160 \pm 3.54\%$  and  $37.744 \pm 4.79\%$  respectively. The cooking loss value obtained in this research was not much different from the value obtained by Kusumastuti (2006) when conducting a study of fattened sheep, that is, 37.17%. The high binding power of the water in meat will lower the cooking loss value. The factors that may affect cooking loss are pH, sarcomere length of muscle fibers, long pieces of muscle fibers, myofibril contraction status, size and weight of the meat samples, and a cross section of the meat. Soeparno (2005) stated that many factors could affect the cooking loss of the meat, among others, the cooking loss can increase when the muscle fibers are shorter. When the water holding capacity of the protein is low, the cooking loss will increase and the tenderness of the meat will reduce.

### **Tenderness**

The results showed that there was a significant difference ( $p < 0.05$ ) between the tenderness of Kacang goat and Garut sheep meat (Table 1). Garut sheep meat is more tender than Kacang goat meat. However, both of them are classified into the category of tender meat. This is consistent with the statement of Suryati (2008) that the tenderness criteria based on the trained panelists showed that very tender meat had a WB (Warner Blatzler) breaking of  $< 3.30 \text{ kg} / \text{cm}^2$ . One of the factors that influence meat tenderness is the cattle age. In this research, the cattle used were less than one year old, which is still relatively young for goat or sheep. The meat of young cattle have more tender compared old cattle.

The factors that affect meat tenderness are the treatments before slaughter, including an intensive raising in an individual stall, resulting in the less movement of livestock compared with when raised in a colony enclosure, or even raise through a grazing system. High-motion activities will increase muscle contraction, decreasing the tenderness of the meat. Carcass handling after slaughter such as withering will reduce the shear force of Warner-Blatzler (WB), thereby increasing tenderness. The tenderness levels of Kacang goat and Garut sheep in this study were categorized as very tender. The tenderness was influenced by the feed used in this study, that is, sorghum grain-based concentrates that contain high carbohydrate and high protein which is an excellent source of energy for livestock. Parakkasi (1999) explains that feed ingredients that serve as an energy source are very efficient for the formation of fat in the body. Lawrie (2003) stated that intramuscular fat tends to dilute the binding woven element in the tendon where the fat is deposited. Both Kacang goat and Garut sheep experienced an increase in fat content during the fattening process which automatically increased their meat tenderness.

### **Meat Color**

Meat color is influenced by the concentration of myoglobin in the meat. The a value of the meat color of Kacang goat was 13.96 and Garut sheep 12.236. This value indicates a positive value, leading to reddish color. Although not significantly different, based on the mean value of a, Kacang goat had redder meat than Garut sheep meat. The degree of redness in meat is affected by the content of myoglobin. The higher the myoglobin content, the redder the meat, and the myoglobin content of meat is affected by a genetic factor related to livestock activity. Goat is a breed of cattle that is more active and mobile than sheep. The higher the cattle activity, the more active the process of glycolysis (the change of glycogen into energy from lactic acid) as more oxygen is required. The b value of both types of livestock did not differ much. The b values of Kacang goat and Garut sheep were 5,2438 and 5,164 respectively, meaning that the meat color was bluish color. The L values of the animals were respectively 45,772 and 43,784, meaning that the meat color was bluish red meat (dark red). However, the meat color of Garut sheep was slightly darker than that of Kacang goat. Purbowati *et al* (2006) reported that the higher the slaughter weight of local sheep, the darker the color in *Longissimus dorsi*, which is dark red.

## Conclusion

The physical quality of the meat of Kacang goat and Garut sheep fed sorghum-based diet have different ( $p < 0.05$ ) pH values and tenderness, but both of them are still at normal pH, and the tenderness is classified as very tender. The values of DMA and cooking loss of both meats were not significantly different. The meat color of Kacang goat and Garut sheep was dark red.

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# Weight Loss of Inter-island Transported Cattle from Kupang Is Reduced by Feeding High Protein-Mineral Mix Block during Quarantine and Sea Transportation

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## Abstract

Total weight lost of inter-island trading cattle during quarantine and transportation from Kupang could reach up to 17.4% due to poor feeding and other transportation-related stresses. This study aimed at assessing liveweight change of inter-island transported cattle given a dry-pressed high protein and mineral mixed feed block (HiPromin). The nutrient content of the feed block was 14.5% crude protein and 11.5 MJ ME/kg DM. Twenty bulls (245±10.5 kg) were chosen from a group of readily exported cattle awaiting for shipment which were kept at quarantine holding ground facilities at Tenau, Kupang, East Nusa Tenggara. The bulls were randomly allotted into control and treated group. The control group received ordinary feed given by cattle traders, while the treated group was offered tested feed block at a rate of 2.5% LW. The experiment lasted for 10 days consisted of 6 days in quarantine, and 4 days during voyage to Samarinda. Independent data comparison test was performed to compare the difference between the two groups. Data showed that control bulls lost weight around 710 g/day whereas treated bulls lost only 100 g/d. Dry matter (DM) and crude protein (CP) intake was very low for control group (27 g/kg W and 0.6 g/kg W<sup>0.75</sup> for DM and CP respectively), but medium for treated group (50.2 g/kg W and 7.5 g/kg W<sup>0.75</sup> for DM and CP respectively). It might be concluded that HiPromin feed block can be used for feeding inter-island traded cattle to avoid weight loss during quarantine and shipment.

**Keywords:** exported cattle, feed block, liveweight loss, growth rate, sea transportation

## Introduction

Every year, more than 60,000 fattened cattle were shipped out of East Nusa Tenggara (NTT) to Java (mainly Jakarta) and Kalimantan island for slaughter. However, these cattle experience a significant weight loss (8.5 – 17.4%) due to transportation-related stresses (Leo Penuet *et al*, 2009). Liveweight loss margin depends on the length of time spent in quarantine and travel to final destination (Lailogo; 1989). The longer the time span, the greater the loss is. As a result, yearly regional economic loss from inter-island cattle trading is around Rp 27 – 92 billion. One key factor known to play an important role in this situation is feeding (quality and quantity). Up to present, inter-island cattle traders rely merely upon rice straw or standing hay as the main feed during the quarantine and transportation (sea and land) of cattle. Cattle can stay on this diet for up to three weeks. To overcome this problem, Mullik *et al* (2013) has developed a dry-pressed high protein-mineral mixed feed block (HiPromin) for inter-island cattle. The present study aimed at evaluating response of inter-island traded cattle to HiPromin offered during quarantine and sea transportation to Samarinda (East Kalimantan).

## Materials and Methods

Twenty bulls (245±10.5 kg) were chosen from a group of readily exported cattle awaiting for shipment which were kept at quarantine holding ground in Tenau, Kupang, NTT. The bulls were randomly allotted equally into control and treated group. The control group received common feedstuff given to cattle such as rice straw, dried native grass, corn stover, banana trunk, and a small proportion of fresh leucaena leaves. Feed offered to control group had protein content of 2.5% and metabolisable energy of 8.1 MJ ME/kg DM. Treated group, on the other hand, was offered a tested feed block that contained 14.5% crude protein, 11.5 MJ ME/kg DM, and 12% total mineral at a rate of 2.5% LW. Tested diet was a dry-pressed high protein-mineral mixed (HiPromin). Detail nutrient composition was described in Mullik *et al* (2013).

Each block weight around 2 kg with a dimension of 20 cm x 15 cm x 15 cm. The experiment lasted for 10 days consisted of 6 days in quarantine, and 4 days during voyage to Samarinda.

There was no feed adaption period since the block is designed to be provided directly to the animal soon after they arrive at quarantine holding ground. Feed block was the only new aspect intervened for the treated group, whereas other management aspects such animal handling, water provision, feeding frequency, feeding time were the same as that normally done for control and all other cattle in the group. Therefore, the feeding time was twice a day while drinking water provision is only once a day when the cattle were in quarantine holding ground, but during the four days voyage to Samarinda, the cattle were only given drinking water one time (on day 3). Therefore, feed intake during sailing from Kupang to Samarinda was very low.

Parameters measured were feed intake, liveweight change (LWC), and liveweight loss. Feed intake was measured by weighing the amount of feed offered and refusal every day for each animal. However, daily measurement could not be done on board of the ship due a very limited space. Therefore, on board feed intake measurement was done daily weighing of the feed given, but feed refusals were only be weighed once after all cattle were unloaded from the ship at Samarinda port. Liveweight of the cattle were only measured twice. The first weighing was done when cattle arrived at quarantine holding ground in Tenua, Kupang, whereas the second measurement was done soon after all cattle arrived at cattle pool in Samarinda. These two weight data were used to calculate LWC and LW loss.

All data were statistically analysed using independent data comparison test to compare the difference between the two groups. Statistical analysis was performed using SPSS v.20 software. Difference was detected at P value of <0.05.

## Results and Discussion

### Nutrient Intake

Total intake for dry matter (DM), organic matter (OM) and crude protein (CP) was highly significantly different for the two groups (Table 1). Intake of DM by control group was only 0.68% LW compared to treated group (1.31% LW). The level of intake for both group was very low. An ideal DM intake should be around 3% LW but these group achieved only 23% and 43% of the ideal level for control and treated group respectively.

Table 1. Intake and liveweight change of inter-island transported cattle given common diet or a high protein-mineral (HiPromin) feed block during quarantine and transportation from Kupang to Samarinda

Parameter	Control	HiPromin	SEM	P value
Dry matter intake:				
% Liveweight	0.68	1.31	0.029	<0.01
Gram <sup>-kg liveweight</sup>	6.8	13.0	0.297	<0.01
Gram <sup>-kg metabolic weight</sup>	26.9	50.2	1.158	<0.01
Organic matter intake:				
% Liveweight	0.60	1.16	0.058	<0.01
Gram <sup>-kg liveweight</sup>	6.0	11.6	0.570	<0.01
Gram <sup>-kg metabolic weight</sup>	24.1	44.6	2.249	<0.01
Crude protein intake:				
% Liveweight	0.01	0.20	0.009	<0.01
Gram <sup>-kg liveweight</sup>	0.15	1.95	0.092	<0.01
Gram <sup>-kg metabolic weight</sup>	0.57	7.51	0.353	<0.01
Liveweight:				
Initial (kg)	250	220		
Final (kg)	243	219		
Total weight lost (g)	7,142	1,571	0.895	<0.01
Percent weight lost	2.89	0.68	0.391	<0.01
Daily weight lost (g)	714	102	53.36	<0.01

Reasons for the low DM intake were slightly different for the two groups. For control group, there were three most probable contributing factors e.i. nutrient deficiency, water inadequacy, and transportation-related stresses. Nutrient deficiency in control cattle related inadequate amount of feed given and poor nutrition of the feed. Our observation on the feeding management revealed that control cattle were offered limited amount of feed every day. For example, a group 3-4 cattle was given only two bunches of feed with a total weight approximately 10-15 kg DM/day. These cattle with an average live weight of 250 kg should be given around 7 kg DM per head/day. In terms of nutrient content, the mixture of the feed given to the control cattle contained only 2.5% crude protein and 8 MJ ME/kg DM since the sole feed given to control animals was dried native grass. In addition to these two factors, the cattle were also exposed to a high stress situation during transportation such as crowdedness, rough sea that might cause sea sickness, and improper handling by technical staff.

For treated cattle, on the other hand, the low DM intake unlikely to be related to nutrient content of the feed offered, rather to inadequate water intake and transportation-related stresses as previously described. It is well documented that dehydration has a tremendous negative effects on feed digestion, nutrient transport and utilization at tissue level, osmotic pressure of the body fluid, and excretion of metabolic wastes (Faries *et al.* 1997; Landefeld and Bettinger 2015). Malloy *et al* (2008) recorded a declined in the intake of dehydrated Zebu cattle by 40%. In a review of literature, Renaudeau *et al* (2012) concluded that heat exposure triggers the internal metabolic system of the animal to make adjustment automatically in the behavior including reducing DM intake.

### **Liveweight Change**

Daily weight loss obtained in the present study was 714 g/d and 102 g/d for control and treated group respectively (Table 1). This data suggested that the HiPromin feed block can be effectively used as the sole feed for inter-island traded cattle as a way of minimizing weight loss during quarantine and transportation to destination cattle pool. Originally, nutrient composition in the HiPromin feed block was designed to support a medium liveweight gain (300 - 400 g/d) if consumed at a level of 3% LW. Unfortunately, low intake achieved in the present study (1.31% LW) has prevented the cattle to perform at the targeted level. Therefore, any management strategies to create a comfortable environment for inter-island traded cattle to achieve their potential intake will have a positive effect on liveweight during quarantine and sea transportation. These include provision of adequate drinking water, proper handling, and reducing on board stock density.

When the cumulative liveweight loss data were calculated as proportion of the LW, the values were 2.89% and 0.68% for control and treated cattle respectively. These data clearly shows that the LW loss of control cattle in the current experiment was far below those values (8.5 – 17.4%) reported by Leo Penu *et al* (2009). It is true that the longer the time spent in quarantine and sea transportation, the greater the loss is. However, the rate of weight loss should be within biological limits. Daily weight loss for control cattle in the present study was already very high for this type of cattle (714 g/d) since the mean LWG for fattened Bali cattle given feedlot diet in NTT is only around 800 g/d (Mullik *et al*, 2014). Therefore, probable explanations for this high weight loss were nutrient deficiency and severe dehydration. Inadequate supply of nutrients from feed into tissue level allowed the body metabolic system to extract nutrients from nutrient deposit in the body causing weight loss. Similarly inadequate external water supply will trigger water depletion from the body (El Nouty *et al*, 1980; Landefeld and Bettinger, 2015; Boyles, 2015). These might also be the reason for the higher weight lost found by Leo Penu *et al* (2009).

### **Conclusion**

It might be concluded that HiPromin feed block can be used as sole feed for inter-island traded cattle to avoid weight loss during quarantine and shipment. However, a proper feeding management need to be adopted to achieve high intake of feed block to ensure minimum or no weight loss.

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**THEME G.**  
**ANIMAL PHYSIOLOGY, BEHAVIOUR, AND WEL-**

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# Level of Malondialdehyde (MDA), Uric Acid and Lymphocyte: Neutrophil Ratio of Laying Hen in The Different Temperature Humidity Index (THI)

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## Abstract

*This study is conducted to investigate the effect of THI on the concentration of MDA, uric acid blood plasma and lymphocytes: heterophil ratio of laying hen. One hundred laying hens are randomly divided into two treatments, each treatment consist 50 birds. Treatments in this work are two levels of temperature humidity index (THI = 89 and 72), which calculated based on dry and wet bulb thermometer in laying housing, located in Kuningan and Bandung. The experiment period lasted 8 weeks, blood samples were collected weekly. Malondialdehyde, uric acid and lymphocytes: heterophil ratio is recorded by spectrophotometer following Biolabo Kit. Data are analysed using paired t-test method. Based on results of this study, the level of MDA and uric acid significantly increase ( $P < 0.01$ ) in the high THI (89). On the other hand, lymphocytes/heterophils (H/L) ratio decreases ( $P < 0.05$ ). These data suggest that high THI (89) contributes of laying stress is caused by the level of temperature and humidity in environment and inside the housing.*

**Keywords:** H/L, laying hen, malondialdehyde, THI, uric acid

## Introduction

Laying hens is the land fowl which has the highest sensitivity towards environmental heat stress, either caused by metabolic heat production or environmental stress in cages (Mushawwir and Latipudin, 2012). The environmental factors which often change are temperature and humidity. The temperature and humidity of environment are able to determine a comfort zone for livestock whose index is usually called by Temperature-Humidity Index (HTI).

High temperature is one of environmental factors which affects to the metabolic profile in cattle's body and causes the heat stress. The heat stress is exacerbated by increased humidity. This condition is indicated by the increase of THI index.

Chicken are homoiotherm animal that always try to maintain their body temperature which one of the ways is by increasing the secretion of glucocorticoids carried out by adrenal cortex and epinephrine by the adrenal medulla (Mushawwir dan Latipudin, 2012). The secretion occurred is due to the increase of adrenocorticotropic hormon (ACTH) that is caused by environmental stress (Wilson *et al.*, 2007; Wheelock *et al.*, 2010; Won *et al.*, 2012).

The increase of glucocorticoids secretion results the gluconeogenesis and turns up the protein metabolism. However, it also decreases the anabolism, so that the synthesis of lymphocytes is decreasing while heterophils increasing. The protein metabolism can form urea. Urea is an end product of protein metabolism in which if the profile in body is too high, it is excreted through the urine. Several amino acids, besides forming the urea, they also can become a precursor of other compounds, for example purine, pyrimidine, and hormones (Mushawwir *et al.*, 2010). Purine and pyrimidine bases are converted into uric acid in the body. Uric acid is an end product or waste product resulted by metabolism/breakdown of purines. The too high-uric acid profile in the body causes pro oxidant and endangers the livestock and causes free radicals.

Free radicals are increasing along with the increase of heat stress as a result of oxidative phosphorylation which produces Reactive Oxygen Species (ROS). The ROS causes the lipid peroxidation rate increases. Product of this oxidation is malondialdehyde (MDA). The levels of ROS are equal to the levels of MDA.

Based on abovementioned descriptions, it is necessary to examine the blood uric acid, the levels of MDA, and the ratio of lymphocyte as a stress indicator due to heat stress for layer-phase laying hens on the different temperature-humidity index (THI).

## Materials and Methods

### Livestock Samples

This research examines 100 laying hens leghorn which are each 50 hens placed in a cage in Kuningan and Cililin (Bandung), in which with the average of Temperature-Humidity Index (THI) is 89 (in Kuningan), and 72 (in Cililin, Bandung). The livestock were kept for two months.

### Blood Collection

The collection of blood sampling is carried out on each chicken samples for two times, which are on the 30<sup>th</sup> day and the 60<sup>th</sup> day. Each blood samples is collected as many as 3 mL using venojet with EDTA.

### Blood Analysis

Analyzing blood aims to know the levels of Melondiadehide (MDA) and uric acid whose process is carried out by following the procedure of KIT-BIOLABO. The recording of MDA and uric acid is conducted by using a spectrophotometer with wavelength of each is 532 nm and 540 nm. The measurement of lymphocytes and heterophils levels is performed by injecting “whole blood” into hematology analyzer Mindray BC-2800.

### Data Analysis

The data obtained (MDA levels, Uric Acid levels, and Lymphocytes ratio: Heterophil) are analyzed by using method of unpaired T-Student with IBM SPSS Statistic 21.

## Results and Discussion

Based on results of the research, the effects of THI towards MDA levels, Uric Acid levels, and Heterophils Lymphocytes ratio is shown on the following Table:

Tabel 1. The MDA levels, Uric Acid levels, and Heterophyl Lymphocytes Ratio of Laying Hens in the locations with the different Temperature Humidity Index (THI)

Blood Indicator	THI		p-value
	89	72	
MDA (nm.M <sup>-1</sup> .cm <sup>-1</sup> )	2.271 <sup>a</sup>	1.055 <sup>b</sup>	< 0.01
Uric Acid (mg.dL <sup>-1</sup> )	7.426 <sup>a</sup>	4.893 <sup>b</sup>	< 0.01
Lymphocytes:Heterophils (ratio)	0.312 <sup>a</sup>	0.152 <sup>b</sup>	< 0.05

Note: The average levels of MDA and Uric Acid is highly significant different ( $p < 0.01$ ), while the ration of Lymphocytes: Heterophils is significantly different ( $p < 0.05$ ).

The result of analysis shows that the uric acid level in the maintenance locations with THI=89 is highly significant different ( $p < 0.01$ ) which is 2.271 mg/dL. It is higher than laying hens at THI=72, which is 1.055 mg/dL. The increase of blood uric acid in layer-phase laying hens is caused by chicken responds to the heat stress by activating the neurogenic system to stimulate Corticotropin Releasing Factor (CRH), so that pituitary anterior issues Adenocorticotropin (ACTH). Furthermore, medulla adrenal secretes the epinephrine which functions to stimulate the second messenger, namely adenylate cyclase (Mushawwir dan Latipudin, 2013). The adenylate cyclase catalyzes the formation of cAMP which moreover, the cAMP will activate the protein kinase A which plays a role in the regulations of metabolic-enzymes and gene transcription, such as triggering glycogenolysis.

The increase of cAMP can improve the uric acid formed. The increase can improve the synthesis of AMP, which is one of purine nucleotide. Moreover, AMP is deaminated to inosine which is later being hydrolyzed to produce hypoxanthine and D-ribose. Furthermore, hypoxanthine is converted to xanthine, and xanthine gets converted to uric acids by xanthine oxidase.

The enzyme Hypoxanthine-guanine phosphor ribosyl transferase (HGPRT) is one of enzyme involved in the reaction of purine bases utilization into nucleotides. This enzyme plays a role in converting purines into purine nucleotides, so it can be reused as a constituent of DNA and RNA. If this enzyme is deficient, purine in the body can be increased for it is not metabolized by enzyme HGPRT which later caused the purine will be metabolized by enzyme xanthine oxireductase (XOR) into uric acid.

The increase of ambient temperature and heat stress on the chickens triggers the increase of enzyme XOR's activity (Donsbough *et al.*, 2010; Mushawwir *et al.*, 2011; Settle *et al.*, 2012). This condition will trigger the increase of uric acid which circulates in blood. In the same condition, it will be known that heat stress decreases the activity of enzyme XOR, so this condition can be ascertained will give impacts towards the increase of uric acid level in blood.

The increase of THI also causes an increase of Reactive Oxygen Species (ROS) which affects to the formation of MDA. MDA levels of laying hens at THI=89 which is 7.426 is highly significant different ( $p<0.01$ ), is higher than laying hens at THI=72 which is 4.893.

ROS is a derivative of oxygen which is more reactive than oxygen in the basic conditions. ROS can get into the blood stream in which if the condition is high prooxidant activity due to free radicals, it often causes oxidative stress. According to Bottje (1995), ROS induces peroxidation of fatty acids with protein, cellular nucleic acids, fats, especially PUFA, hence resulting lipid peroxidation. The main target of lipid peroxidation by ROS is PUFA in membrane lipids. The PUFA degraded by ROS will cause the form of aldehyde like MDA. MDA is a highly reactive three carbon dialdehyde which can be obtained from hydrolysis of pentose, deoxyribose, hexoses, amino acids, and DNA (Evans, 1991; Bottje *et al.*, 1995).

The increase of MDA as an impact of increasing THI shows that the increase of environmental stress is a combination of temperature and humidity. The high environmental stress leads the trajectories of catabolism (Mushawwir *et al.*, 2010) and anabolism (Hardy *et al.*, 2005; Favlik *et al.*, 2007) are activated at the same time, in order to homeostasis defends the normal physiological conditions and the provision of energy.

The increase of free radicals also can be triggered by the rising of trajectory of glycolysis and glycogenolysis, and also oxidative phosphorylation in mitochondria matrix for ATP biosynthesis. The concentration of free radicals (ROS) becomes a key trigger for formation of MDA.

The high concentration of ROS results a reaction with fat, protein, cellular nucleic acids. It then causes a local damage and particular organ dysfunction. Fats are biomolecules that are susceptible towards free radical attack. The component of animal cell membranes contains many sources of PolyUnsaturated Fatty Acid (PUFA). Uppu (2010) in line with Mushawwir and Latipudin (2012) state that PUFA is biomolecules which easily damaged by oxidizing substances whose process commonly called by lipid peroxidation. Evans (1991) reports that breaking the bonds of carbon during lipid peroxidation leads to the formation of aldehyde, such as malondialdehyde (MDA).

Based on the hormonal perspective, heat stress caused by highly THI (89) causes the secretion of Adrenocortico Tropic hormon (ACTH) becomes increased, thus it stimulates the secretion of hormone steroid, namely cortisol. This cortisol obstruct the anabolism which affects to the decrease of lymphosit (L) and the increase of heterophils level (H). So that, the ratio of H/N is higher ( $p<0.05$ ) at THI = 89 which is 0.312, than at THI = 72 which is 0.152.

The high temperature with highly environmental humidity can worsen the condition of heat stress (Marai dan Haebe, 2010). In addition, Marai and Haebe (2010) state that level of cortisol will increase during acute heat stress condition. This is caused by glucocorticoids can lead to the transport of amino acids and fats from cells. So, it can be used directly as a source of energy and for synthesis of other compounds including glucose which is needed by various tissues of the body (gluconeogenesis).

The high cortisol can cause damage of lymphoid glands (thymus) and extension of heterophils' age. This change also results an increased number of heterophils in blood circulation (Kim *et al.*, 2006; Ogura *et al.*, 2007). The results of previous studies as reported by Zapata *et al.*, (2004) and Milne *et al.*, (2012) show that the increase of glucocorticoid (cortisol) can reduce the secretion of cytokines IL-2 which leads to the decrease of lymphocytes proliferation and makes cells of lymphocytes become delicate towards apoptosis. There by it reduces the amount in circulation.

## Conclusion

The high Temperature-Humidity (THI) index up to 80 leads to the increase of MDA and Uric Acid levels, and also the decrease of the ratio of lymphocytes to heterophils. This condition shows that index of THI up to 89 can cause a heat stress for layer-phase laying hens.

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# The Using of Thermograph as Non-Invasive Method to Observe Subclinical Mastitis in Tropical Dairy Cattle

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## Abstract

Temperature is a prominent indicator in physiological status of dairy cattle since heat stress is highly affected in tropical dairy cattle. The changes in heat emitted from udder might be used as presumption of subclinical mastitis. The aim of this research was to observe the variation of udder warmth by using non-invasive method-infrared technology measurement, thermograph and linked it up to subclinical mastitis. This research was conducted in Kunak dairy farming, located in Bogor, Indonesia during January to April 2015. Totally, 53 milk samples from 30 dairy cattle were used in normal lactation stage. Subclinical mastitis was assessed by IPB 1 Mastitis test during milking time. However, the increment of udder temperature then compared with the diagnostic value of IPB 1 mastitis test and somatic cell count. The finding of this research reflected significant increment of udder temperature on lactation dairy cattle. Udder temperature rose dramatically from 33.2 °C to 37.64°C in different range of diagnostic value of IPB 1 mastitis kit; weak (+), distinct positive (++) and strong positive (+++). Several bacteria also confirmed in overall mastitis data observation, i.e. *Staphylococcus* sp, *Streptococcus* sp and, *Corynebacterium*. The levels of somatic cell count were strongly associated with the number of mastitis level. Overall observation by using thermograph was able to detect the fluctuation of udder temperature in tropical dairy cattle which indicated the detection of subclinical mastitis.

Keywords: dairy cattle, subclinical mastitis, thermograph,

## Introduction

The tropical dairy cattle has mainly problem in heat stress, due to the high environment temperature. However, the thermal environment is a major factor that can negatively affect milk production of dairy cows, especially in animals of high genetically potentiation. In addition, the dairy physiological status might be measured from body temperature emitted. It's well known that temperature is the prominent indicator of dairy physiological status. The radiation temperature estimation is usually performed through the infrared radiation intensity measurements. The source of this radiation is the body surface, so the measured values describe first of all the skin temperature of the animal. The use of Infrared Thermography has proved to be an interesting and non-invasive tool in veterinary research, since it is highly sensitive for detecting temperature changes of skin surface (Scoot *et al.*, 2000; Hovinen *et al.*, 2008). This technique involves the relationship between the variation in subcutaneous blood flow and the heat emission from the studied area, detected by the thermograph in a colorimetric scale correlated with changes in temperature (in Celsius degree).

Mastitis is one of infectious diseases that affect dairy herds and causes great damage to the milk productive chain. Mastitis is an udder inflammation that typically results in temperature rises of the affected area, followed by a reduction in milk secretion and changing in permeability of the membrane which separates the milk from the blood (Bramley *et al.*, 1996). Hovinen *et al.* (2008) showed the capacity of the infrared thermography in the identification of temperature increases (> 1°C) in cow's udder with the clinical mastitis. Besides the infrared thermography does not cause damage to animal's health, the technique is able to evaluate the welfare in different environmental conditions (Colak *et al.*, 2008). The aim of this research was to observe the variation of udder warmth by using non-invasive method-infrared technology measurement, thermograph and linked it up to subclinical mastitis

## Materials and Methods

This research was conducted in dairy farming centre, Kunak, Bogor, West Java during January to

April 2015. As totally 30 dairy cattle were used in this research in normal lactation stage. Furthermore, the mastitis clinical test was measured by IPB 1 mastitis test during milking time.

Subclinical mastitis test was tested in milk from each part of udder by using IPB-1 reagent. For 2 ml of milk sample was poured into the mastitis kit and added IPB-1 reagent equally. It was shake horizontally for 15-20 second. After that the respond of the test may seen directly. We compared the mastitis test with its standard (-, +, ++,+++).

Based on subclinical mastitis method were used previously, we then compared it into microbial test status (patogen bacteria) by using blood agar base. Milk sample from each udder compartment were collected hygienically and collected in the sterile tube. Inoculation process was applied on blood agar baseto distinguish patogen bacteria.

Somatic Cell Count (SCC) was measured by Breed method (Sudarwanto, 2012). At first time, milk sample was homogenized and several drops were obtained by using Breed pipet. Drop the milk sample for 0.01 ml into object glass (1 cm<sup>2</sup>). Make it flat and thin, then dry it for 5-10 minutes. The calculation of SCC was counted under Microsoft condition. However, this calculation may be able to describe the amount of somatic cell in milk sample.

Measurement udder thermal was conducted using thermograph. Cattle placed in a cage and a thermal camera was recording udder around 2-3 minutes. A distance from cattle to thermal camera was about 1-2 meter approximately.

## Results and Discussion

The principle of infrared camera, thermograph is that all objects emit infrared radiation proportional to their temperature according to the Stefan-Boltzmann law via conduction, convec- tion, and radiation. A thermal camera absorbs infrared radiation and generates pictorial images based on the amount of heat generated, without causing radiation exposure (Eddy *et al.*, 2001; Mazur and Eugeniusz-Herbut, 2006; Kunc *et al.*, 2007). In more applied science, recently it may be used in agriculture sector especially for livestock behaviour and physiology. This technology may implement for animal welfare and health status since it's non-invassive, not disturbing the animal directly.

In this study, this camera was used to record several data observations for udder thermal to detect sub-clinical mastitis case. It could be seen there's a trend of the increment udder thermal and the amount of somatic cell count (Table 1).

Table 1. The average of somatic cell count and udder warmth temperature by using thermograph

No	IPB-1 Value Diagnostic Test	Somatic Cell Count (Log/ml)	Udder Warmth Temperature
1	+ (Weak)	5.202±0.511	33.2 ± 2.65
2	++ (Distinct Positive)	5.754± 0.634	36.2 ± 1.30
3	+++ (Strong Positive)	6.463±0.549	37.64± 0.56

Table 1 presented the variation of udder temperature observed by using thermograph. It can be seen that when temperature was only 33.2°C the diagnostic value of IPB-1 test was (+) or weak with total of somatic cell count was around 5.202 Log/ml. However, udder temperature was significantly increase or when temperature reached 36,2 °C with diagnostic value distinct positive (++) with the total of somatic cell count was 5.754Log/ml. When temperature reach more than 37.64 °C the diagnostic value of IPB-1 test kit showed strongly positive (+++) with the average of somatic cell count was 6.463 Log/ml. This argument was quite similar to Mellenberger *et.al*(2014) which stated that strongly positive in mastitis may seen when the somatic cell count was reach more than 6.7 Log/ml (5.000.000 cell count/ml).

Pathogen bacteria examination was conducted to strengthen the result of indirect measurement by using IPB-1 test during detecting udder condition in each udder compartment. The test of pathogen bacteria detected several bacteria such as NCS (*Negative Coagulation Staphylococcus*), *Staphylococcus sp*, *Streptococcus sp*, *corynebactrium*, mix pathogen and fungi. Based on 53 sample, it was found the existing of *Staphylocococcus sp* and *Corynebactriun* for 26%, NCS and coliform for 13% respectively, *Streptococcus sp* for 9%, and the rest were mix and fungi for 13%.

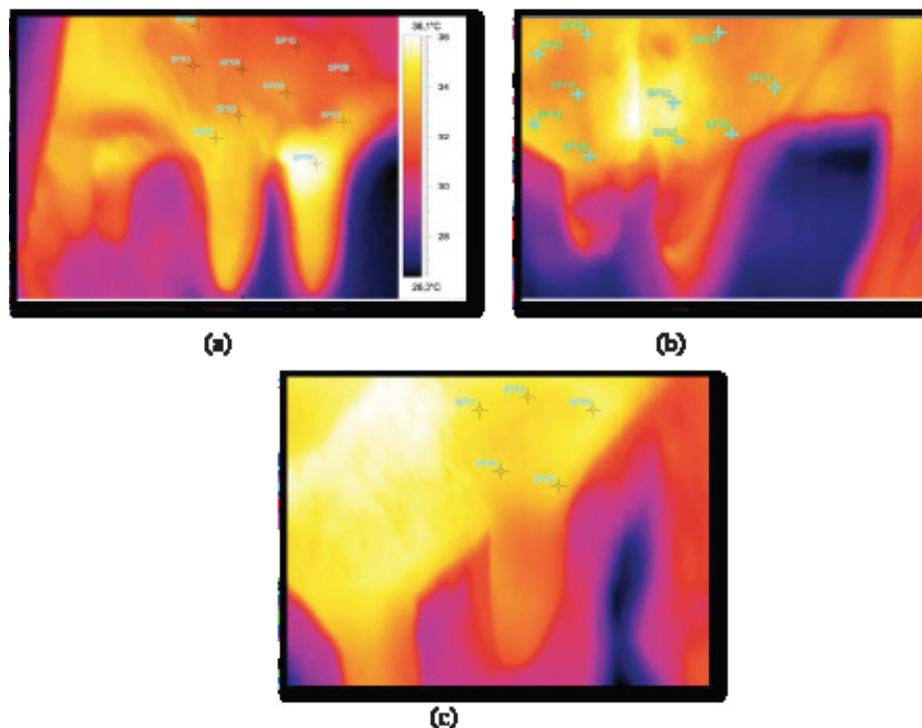


Figure 1. Udder temperature appearance recorded by Thermograph (a) weak in mastitis (+), (b) distinct positive (++), (c) strong positive (+++)

As stated previously, subclinical mastitis is the udder inflammation case. However, the diagnose test was emphasized by the amount of somatic cell count (SCC) and the existing of pathogen bacteria. The pathogen comes into the udder by the teeth pathway and growth well in udder. This situation will result in biological and metabolic product. These materials will irritate the tissues and raise the inflammation. The declining of milk production and increasing of SCC were highly connected to inflammation. The somatic cell is the immune respond due to the inflammation process.

## Conclusion

In this study, thermograph showed that there was a slightly increment of udder temperature in different sub-clinical mastitis condition. This information is highly useful to detect sub-clinical mastitis disease with non-invasive method for animal welfare. However, we need more data to have more persisted information about specific temperature when the value of diagnostic sub-clinical mastitis is declining/inclining.

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# Physiological Response and Blood Profile of Sheep Reared in Petir Village and Fed Cassava Tops Silage (*Manihot esculenta* sp.)

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## Abstract

*Under traditional system the farmer fed their sheep without considering the quality of feed given. Cassava leaves reach in protein found around the village can be used to improve the quality of feed. This study aimed to examine the effect of supplementation of cassava leaves silage or concentrate into commonly given forage on physiological responses and blood profile of sheep. This study was carried out in Petir village using sheep kept by the farmer under traditional rearing system. Completely randomized block design with 4 treatments and 4 replicates was used. The treatments were T0 (100% unchopped forage), T1 (100% chopped forage), T2 (80% chopped forage + 20% concentrate), and T3 (80% chopped forage + 20% cassava leaves silage). The variables observed were respiration rate, heart rate, rectal temperature, haemoglobin, hematocrit, erythrocytes, and leucocyte. Data were analyzed using analysis of variance (ANOVA). Any means differences were further tested using Duncan Multiple Range Test. The results showed that the treatments had no effects on rectal temperature and heart rate. Respiration rates measured in the morning and at noon of sheep in T2 and T3 groups were higher ( $P < 0.05$ ) than those of sheep in T1 group. Heart rate measured at noon of sheep in T3 group were higher than those of other groups. The treatments had no effects on blood profile except on white blood cell. Sheep in T2 group had higher ( $P < 0.05$ ) leukocytes than those of other groups. It is concluded that cassava leaves silage or concentrate diets caused high heat production of sheep but it can be countered by better heat dissipation.*

*Keywords: sheep, cassava leaves silage, physiological responses, blood profile*

## Introduction

Sheep farming in Indonesia is largely operate in subsistence small scale. Technology is not applied even a simple one such as chopping the forage so that it can be consumed with little waste. The main problem faced by farmers today is the low quality and scarcity of forage especially during dry season. Another problem faced by farmers is the limited ability of farmers in the supply of concentrate feed due to the concentrate price is high. This leads to low productivity of sheep. Therefore it is necessary to find out alternative feed to supplement grass so as to improve the quality of feed and finally optimizing the productivity of sheep. One alternative feed that can be used to supplement the grass is cassava leaves (*Manihot esculenta* sp.). Cassava leaves has a crude protein content of 21-24 % (Sokerya & Preston 2003). However, its high HCN content become a constraints in its use. Making cassava leaf silage may solve the problem. Silage has long shelf life and can reduce levels of HCN content by 78.67 % (Noveanto 2013 ). The aim of this study, therefore, was to evaluate the effect of cutting forage supplemented with cassava leaf silage or concentrate on the physiological response and the blood profile of sheep reared by the farmer in Petir village, Bogor, west java.

## Materials and Methods

Sixteen local male sheep with average initial body weight of  $21.55 \pm 2.02$  kg were placed in individual cages. The cage was provided with feed trough and bucket for drinking water. They were fed experimental diet consisted of leaves of sweet potato (*Ipomoea batatas*), cassava leaves silage (*Manihot esculenta* sp.) and concentrate which composed of coconut cake, peanut meal, wheat pollard, milled cassava,  $\text{CaCO}_3$ , salt and premix. The experimental design used was a randomized block design with 4 treatments rations and 4 groups (by weight). The treatments were as follows :

T0 : 100 % forage is not in the chop

T1 : 100 % forage in the chop

T2 : 80 % forage in chop + 20 % concentrate  
 T3 : 80 % forage in chop + 20 % cassava leaf silage

Data were analyzed using analysis of variance (ANOVA), and any significant differences of treatment means was followed by Duncan multiple range test (Steel and Torrie 1993). Variables measured were rectal temperature, respiratory rate, heart rate, hematocrit, hemoglobin, erythrocytes and leucocytes.

## Results and Discussion

Results showed that cutting forages with cassava leaves silage or concentrate supplementation did not significantly affect either the sheep rectal temperature in the morning, afternoon, or evening (Table 1). Mean daily rectal temperatures in sheep studies ranged 38.71-39.10 °C. Sheep rectal temperature in this experiment was still in the normal range. Sonjaya (2012) shows that the lambs rectal temperature under normal conditions varied between 37.90 - 39.80 °C.

The differences are not apparent during the study showed that the sheep are less affected by the treatment of feed. Isnaeni (2006) mentions that the body temperature of most animals are more affected by ambient temperature. Sudarman and Ito (2000) reported that the sheep were given different protein levels showed significant differences for vaginal temperature at 20 °C but no real effect on the temperature of 30 °C.

Table 1. Physiological responses of sheep fed different physical form and supplementation of feed

Parameters	Treatments*)	Time measurement		
		Morning	Noon	Afternoon
Rectal temperature (°C)	T0	38.57 ± 0.48	39.11 ± 0.32	39.31 ± 0.25
	T1	38.33 ± 0.32	38.77 ± 0.28	39.02 ± 0.84
	T2	38.76 ± 0.52	39.17 ± 0.38	39.37 ± 0.36
	T3	38.60 ± 0.35	39.12 ± 0.24	39.22 ± 0.27
Respiration rate (resp/min)	T0	30.38 ± 7.04 <sup>ab</sup>	59.85 ± 11.54 <sup>ab</sup>	62.80 ± 22.64
	T1	28.20 ± 4.05 <sup>a</sup>	56.55 ± 9.84 <sup>a</sup>	54.78 ± 20.68
	T2	36.15 ± 8.39 <sup>b</sup>	77.40 ± 24.99 <sup>bc</sup>	69.23 ± 28.32
	T3	34.88 ± 7.42 <sup>b</sup>	80.70 ± 23.17 <sup>c</sup>	70.10 ± 29.93
Heart rate (beat/min)	T0	75.78 ± 9.97	87.31 ± 11.58 <sup>a</sup>	98.78 ± 13.44
	T1	78.33 ± 12.65	90.78 ± 14.78 <sup>a</sup>	99.43 ± 16.68
	T2	85.69 ± 12.14	98.71 ± 14.43 <sup>ab</sup>	101.80 ± 16.25
	T3	86.39 ± 12.60	104.13 ± 11.07 <sup>b</sup>	102.38 ± 14.42

Different superscript within the same column differ significantly (P<0.05). T0 : 100 % Forage is not in the chop, T1 : 100 % forage in the chop, T2 : 80 % forage in chop + 20 % concentrate, T3 : 80 % forage in chop + 20 % cassava leaf silage

The cut forage with supplementation of cassava leaf silage or concentrate significantly (P<0.05) affect the frequency of respiration sheep in the morning and afternoon. The average of the highest frequency indicated by sheep with cassava leaf silage supplementation treatment (T3) of 61.89 ± 29.57 times/min and the lowest is shown by sheep by administering 100% forage in chop (T1) of 46.51 ± 18.62 times/min. The high frequency of respiration in sheep T3 because of the high content of protein in the ration T3 (19.84%). High consumption of nutrients that will increase the body's metabolic processes of the body so that the heat generated will be more (Wuryanto *et al.* 2010). Isnaeni (2006) adds that at the time of increased metabolic rate, oxygen consumption and carbon dioxide formation also increased. So the sheep will increase the frequency of respiration to reduces body heat received and meet the need of oxygen. Sheep respiration frequency of the lowest found in sheep T1 is equal to 46.51 ± 18.62 times/min. Cutting forage becomes shorter size can decrease the processes of digestion and rumination so that reduce heat increment.

The treatment significantly affected (P<0.05) heart rate of sheep during the day. The average heart rate during the study ranged 87.29-97.63 sheep beats/min. The average heart rate is still within the normal range. This is in accordance with Frandson (1992) who reported that normal heart rate of sheep in tropical regions ranged between 60-120 beats/min. Sheep fed cassava leaf silage had the highest heart rate (104.13 ± 11:07 beats/ min). This may be caused by different dry matter intake at each treatment. Lamb in T3 treatment had higher DM intake (813 ± 86.64 grams of tail-1 day-1) than the other treatments T0 (810 ±

86.10 grams of tail-1 day-1), T1 ( 769 ± 78.65 grams of tail-1 day-1), and T2 (775 ± 167.44 grams of tail-1 day-1). High dry matter intake improved feeding activity that increases the heart rate.

Results of blood profile showed that feed treatment had no significant difference on the number of sheep erythrocytes (Table 2). The average number of sheep erythrocytes in research ranging from 8.53-8.60 million/mm<sup>3</sup>. This figure is higher compare to that of Astuti *et al.* (2008) who reported that the sheep were kept in the Forest Education Gunung Walat has a had number of erythrocytes of 7.85 million/mm<sup>3</sup> and 5.74 million/mm<sup>3</sup> on the young and adult sheep, respectively.

Tabel 2. Blood profile of sheep fed different physical form and supplementation of feed

Parameters	Treatments			
	T0	T1	T2	T3
Erythrocyte (millions/mm <sup>3</sup> )	8.53 ± 0.82	8.60 ± 0.92	8.04 ± 1.47	8.47 ± 1.16
Haemoglobin (g/dL)	7.85 ± 1.23	7.08 ± 1.04	7.05 ± 1.34	7.80 ± 0.28
Haematocryte (%)	31.75 ± 1.89	28.25 ± 3.86	29.50 ± 3.11	30.50 ± 1.00
Leucocytes (thousand/mm <sup>3</sup> )	5.11 ± 0.36 <sup>a</sup>	6.51 ± 0.42 <sup>b</sup>	5.71 ± 1.03 <sup>a</sup>	5.58 ± 0.53 <sup>a</sup>

Different superscript within the same row differ significantly (P<0.05). T0 : 100 % Forage is not in the chop, T1 : 100 % forage in the chop, T2 : 80 % forage in chop + 20 % concentrate, T3 : 80 % forage in chop + 20 % cassava leaf silage

Treatments did not significantly affect hemoglobin levels of sheep. Mean hemoglobin levels ranging from 7.05 - 7.85 g/dL. The results are below normal hemoglobin levels in sheep (Pugh 2002) range from 9 - 15 g/dL. Low hemoglobin level is proportional to the number of sheep erythrocytes result.

Hematocrit of sheep did not affected by the treatment. The mean hematocrit values ranged 28.25 - 31.75 %. The results are below the normal range sheep hematocrit value reported by Smith and Mangkiwidjojo (1998) ranged 32 % -45 %. The low hematocrit value was higly correlated to low red cell count .

Treatment had significant effect (P <0.05) on the number of leukocytes. The number of leukocytes in this study is still in the normal range. Pugh (2002) stated that the number of leukocytes in normal sheep ranges from 4-12 thousand/mm<sup>3</sup>. The results of the study indicate that the sheep were healthy. Leukocyte count of T1 group was higher than that of sheep in T0, T2, and T3. Another possible factor could cause the number of leukocytes in T1 is higher due to saponins in the green leaves of sweet potato. Saponins can act as an imunostimulator that can improve the function and activity of the immune system so that it can boost the immune system (Francis *et al.* 2002).

## Conclusion

Cutting ( chopping ) forage reduce the frequency of respiration and heart rate sheep. Supplementation of cassava leaf silage in the diet increases the frequency of respiration and heart rate sheep. Increased frequency of respiration and heart rate is still within normal limits. Silage cassava leaves can be used as forage supplementation. Cutting forage silage with cassava leaf supplementation did not affect the number of erythrocytes, hemoglobin and hematocrit sheep. Leukocyte count increased in the administration of 100 sheep forage is cut

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**THEME H**  
**ANIMAL ENVIRONMENT MANAGEMENT**

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**Proceeding of the 3<sup>rd</sup> International Seminar on Animal Industry,  
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# N<sub>2</sub>O (Nitrous oxide) Gases Production from Lactating Dairy Cow Feces in Different Management Feeding System

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## Abstract

Lactating dairy cow feces produce N<sub>2</sub>O gases that contribute to global warming. N<sub>2</sub>O is one of greenhouse gasses which is resulted during storage, processing, stacking or deposition and highly influenced by the amount of feces and the type of livestock (IPCC 2012). The aim of this research was to analyze the N<sub>2</sub>O (nitrous oxide) production from lactating dairy cow feces that fed with elephant grass and rice straw. This research was conducted in Kunak Kunak dairy farming, located in Bogor, Indonesia from September-December 2014. Totally 12 dairy cattle were used during this research. N<sub>2</sub>O gases was measured by using gas chromatography (GC). Feces were collected as 1 kg in 24 hours and incubated for 8 weeks to measure N<sub>2</sub>O gases. Randomize complete design was used with here were 3 feeding treatments in this research, RS (100% rice straw+ concentrate+ tofu waste by product), EG (100% elephant grass+ concentrate+ tofu waste by product) and RSEG (50% elephant grass + 50% rice straw+ concentrate + tofu waste by product). Several variables were observed; N<sub>2</sub>O gases, proximate feed analysis, feces proximate analysis, total of feces organic matter, C organic feces, N feces, ammonia (NH<sub>3</sub>) concentration. Based on the research generated N<sub>2</sub>O gases production from lactating dairy cow feces had significant different feeding management system ( $p < 0.05$ ). It was found N<sub>2</sub>O production was found highest in RS compared to EG and RSEG. The incubation process for 8 weeks found there were significantly decreasing amount of feces's substrate and organic matters. The declining of feces's organic matter during fermentation was followed by N<sub>2</sub>O gases production on 8th week in all treatments. It can be concluded that N<sub>2</sub>O gases from lactating dairy cow feces produce higher in RS treatment with highest production on 3th week.

Keywords: feces, lactating dairy cows, N<sub>2</sub>O

## Introduction

Livestock sector is often responsible to the emission of Green House Gasses (GHG) in the atmosphere. The dairy cattle farming system also contribute to potential emission of GHG. The main product of dairy cattle is milk, while the waste such as urine and feces could not be avoided in dairy farming activities. It has been calculated that the average of feces of Frisien Holstein (FH) breed is around 18.45-36.9 kg per day from the animal body weight in average as 225-450 kg (Glenn 1985). As the higher feces produces the more emission might be occurring.

Dairy cattle's feces result in N<sub>2</sub>O which come from livestock sector and contribute for almost 30%-50% of total global emission (Oenema *et al.* 2005). According to IPCC, either solid or liquid waste have equal N<sub>2</sub>O emission during storage, processing, accumulating, and sedimentation that are highly influenced by the amount of feces and feed varieties given. As 90% of dairy cattle farming in Indonesia run by traditional way. Farmers frequently deliver feed in low quality such as Elephant grasses and rice straw which potentially has a higher N<sub>2</sub>O emission. The aim of this research was to analyze the production of N<sub>2</sub>O gasses from dairy cattle feces with different feed source.

## Methods

The feces from 12 lactation dairy cattle were used in this research. Feed source were used such as elephant grass, rice straw, concentrate, tofu waste byproduct, and water. Tools were required such as thermometer, alcohol, container, feces incubator stuffs., syringe BD (Becton Dickinson) label for 10 ml, vacuum blood 10 ml, aluminum foil, plastic wrap, plastic bag, digital measurement for 1000 g, digital measurement for 50 kg. Chromatography Gasses (CG) tool was used in this research.

This research was conducted from September to December 2014 in dairy farming center, and laboratory of waste management processing, Bogor Agriculture University. The dairy cattle were used should be

measured firstly for its weight based on estimation of *Schrool* method. Feed were given for almost 3.5% of dry matter from animal weight. Then, feces resulted for 24 hours were collected and measured in each cattle. In addition, as 1 kg feces sample were taken for incubation process for 8 weeks. There were 3 feeding treatments in this research, RS (100% rice straw+ concentrate+ tofu waste by product), EG (100% elephant grass+ concentrate+ tofu waste by product) and RSEG (50% elephant grass + 50% rice straw+ concentrate + tofu waste by product). Variable observed (N<sub>2</sub>O production, proximate analysis, Nitrogen analyses. The data were analyzed by randomize complete analyses.

## Results and Discussion

Based on proximate analysis, the dry matter in elephant grass was lower (25.47%) compared to rice straw (38.74%) and concentrate (87.58%). Tofu waste by product has 20.83% of totally dry matter (table1). According to Mc.Donald*et.al* (1998), elephant grass has a high water content (81.50%). While Doyle *et.al* (1986) stated that rice straw has a low protein content (2.2%-9.5%).

Table 1. Nutrient content on feed stuff during research

Feed stuff	Water content	Dry matter	Ash	Fat	Protein	Fiber	BETN
	(%)						
Elephant grass	74.53	25.47	3.85	0.70	2.61	6.52	11.79
Rice straw	61.26	38.74	7.10	0.79	1.86	10.72	18.27
Tofu waste by product	79.17	20.83	0.32	0.73	2.31	2.20	15.27
Concentrate	12.42	87.58	22.03	4.43	6.31	13.18	41.63

In fact, the nutrient content in feed was highly connected with N<sub>2</sub>O gasses production. Nutrient composition in feed was the best media growth for the bacteria in resulting N<sub>2</sub>O. Based on data observation, it could be seen, the protein and fiber content in feces before and after incubation time was higher in RG, while the declining of fiber content and organic material after incubation was higher in JP and JPRG (Table 2). This data indicated there was a fast and high degradation process during incubation process.

Philippe and Nicks (2014) stated that the GHG resulted as the degradation of organic material in feces by anaerobic way. Higher fiber content will increase GHG production. Based on data observation, the protein content and fiber content before and after incubation were find highest in EG treatment, but the declining of fiber and organic material after incubation were detected in RS and RSEG (Table 2). The fast declining of fiber content indicated the fast degradation process during incubation. Makkaret.*al* (2007) stated that feed component, mostly fiber and protein contributed to gas production during fermentation. The degradation process could be seen from the declining of feces composition, with the highest proportion JPRG 16.82 g, JP 16.77 g and RG 6.95 g, while protein degradation was followed by JP 6.58 g, JPRG 5.04 g and RG 4.76 g.

Gas production in N<sub>2</sub>O was resulted from nitrification and denitrification process (Bremner danBlackmer1978) and it found mostly in fresh feces in ammonium form (Sommer *et al.* 1992) The data of N<sub>2</sub>O gasses production could be seen in Table 3. There was significantly different (P<0.05) in N<sub>2</sub>O gas production from each treatments in second, third, and forth week of incubation time. In addition, there was not significantly different in first, fifth and sixth of total gas production (P>0.05).

Table 2. Proximate analysis in fresh dairy feces, before and after incubation

Treatments	Feces composition (g)	Before incubation (g)	After incubation (g)	Decreasing composition (g)	Percentage of decreasing composition (%)
RS	Water content	798.40	707.03	91.37	11.44
	Dry matter	201.60	142.97	58.63	29.08
	Ash	51.90	43.10	8.81	16.97
	Fat	4.00	1.87	2.13	53.25
	Protein	18.90	12.33	6.58	34.79
	Fiber	42.00	25.33	16.77	39.83
	BETN	84.70	60.34	24.36	28.76
EG	Water content	830.50	716.21	114.29	13.76
	Dry matter	182.30	133.79	48.51	26.61
	Ash	39.40	23.38	16.03	40.67
	Fat	2.90	2.30	0.61	20.86
	Protein	18.70	13.94	4.76	25.45
	Fiber	43.50	36.55	6.95	15.98
	BETN	77.80	57.62	20.18	25.94
RSEG	Water content	787.90	710.94	76.96	9.77
	Dry matter	212.10	139.06	73.04	34.44
	Ash	49.40	40.97	8.43	17.06
	Fat	3.90	1.70	2.20	56.41
	Protein	16.60	11.56	5.04	30.36
	Fiber	41.30	24.48	16.82	40.73
	BETN	100.90	60.35	40.55	40.19

Table 3. N<sub>2</sub>O gas production from lactation dairy cattle during research

Week	The average of N <sub>2</sub> O gas (ppb)		
	JP	RG	JPRG
1	672.88±87.93	612.13±130.54	686.50±73.68
2	771.38±194.17 <sup>a</sup>	490.75±64.38 <sup>b</sup>	547.00±47.66 <sup>b</sup>
3	1 280.63±531.14 <sup>a</sup>	874.63±284.83 <sup>b</sup>	499.63±74.54 <sup>c</sup>
4	1 052.13±255.20 <sup>a</sup>	782.00±160.65 <sup>b</sup>	739.38±157.60 <sup>b</sup>
5	823.13±122.22	721.63±44.83	847.63±39.08
6	420.63±14.64	407.88±9.45	413.50±18.39
7	ND	ND	ND
8	ND	ND	ND
Total	20 083.00±311.92 <sup>a</sup>	15 556.00±203.73 <sup>b</sup>	14 934.50±160.30 <sup>b</sup>

Information: \* different subscript in the same line shows significantly different (P<0.0%), ND (not detected).

The N<sub>2</sub>O gas production connected to the Nitrogen (N) content in feces. As the faster of declining N content has a high potential in resulting N<sub>2</sub>O. The N content was the source of N<sub>2</sub>O gas production from denitrification process (Chadwick *et al.* 2011). Based on Philippe and Nicks (2014), the N<sub>2</sub>O gas production was occurring as the microbial process from ammonium oxidation. Fermentation process appeared in anaerobic condition result the low production of N<sub>2</sub>O. Bamualimet *al.* (2008) stated that the storage during anaerobic condition was able to decreasing the N<sub>2</sub>O gas production. The highest N<sub>2</sub>O gas was found in JP treatments, proofed by the high degradation N content during fermentation. The declining of N during fermentation was 39.17% JP, 30.61% RG, and 37.17% JPRG respectively (Table 4).

Tabel 4. Nitrogen content in lactation dairy cattle before and after incubation

N content	Perlakuan		
	RS	EG	RSEG
Before incubation (g)	147.18±38.80 <sup>a</sup>	189.38±20.53 <sup>b</sup>	150.50±24.26 <sup>a</sup>
After incubation (g)	89.53±6.47 <sup>a</sup>	131.41±15.69 <sup>b</sup>	94.56±2.28 <sup>a</sup>
N declining percentage (%)	39.17±41.18	30.61±29.00	37.17±23.84

## Conclusion

The N<sub>2</sub>O gas production from dairy cattle feces was higher in rice straw treatments with the peak of gas production in the third week of incubation. During 8 weeks incubation time presented the declining of N<sub>2</sub>O gas, nitrogen and nutrient content in feces.

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**THEME I.**  
**SOCIAL ECONOMY AND POLICY IN ANIMAL PRODUCTION**

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# Estimated Value of Live Buffalo Prices in the Economic Analysis of the Income of Farmers in the Village

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## Abstract

*This research was conducted in the Kuripan District, West Lombok, West Nusa Tenggara Indonesia. The purpose of this study to find out the estimated value of the selling price of live buffalo, bodyweight and price of meat in the economic analysis income of farmers in the village. The research was conducted in 2014 using the method simple random field survey on 23 respondents. Primary data obtained from the respondents, and secondary data obtained from the relevant agencies. Parameter observed were the sale price of live buffalo by using the financial economy analysis of some factors. The factors were the estimated the body weight and prices, body length (Pt), bodyweight (Bk), chest girth (Ld), the price of meat (Hdk). The data were analyzed by using descriptive methods. The results showed that the highest sale value on male buffaloes around Rp.17.09 million, the value of the price of meat at the present around Rp.89.500,-. As for the measurement, calves were not calculated the value of body length and chest girth, because it can not be used for the production of meat, judging by the weight of livestock.*

**Keywords:** buffalo, estimated value, price, revenue analysis

## Introduction

Agriculture is until now, still the mainstay of major as the dominant livelihood, the farmers in the district Kuripan Wes Lombok Wes Nusa Tenggara Province of Indonesia. Important role in farmers managing agricultural land and k buffalo socially ownership can priode special meaning for farmers. The implementation however, in of the number of buffalo are reared by the family scale are still in the possession of approximately 1-3 individuals/farmers, basically livestock as sideline business, as saving time spere and labor processing utilization fileds.

Which can a process in as independent farmers, can eventually generate better profits. Kuripan sub-districts of West Lombok Regency, it is suitable in conditions of land agricultural is seen to be combined buffalo with efforts, known adaptable to the various conditions of the rural agro-ecosystems, and is a complementary effort. Faced obstacle often by farmers is holding that buffalo had mated not could another in farers resulting feel the loss, profit because kept animals not making.

The District of West Lombok Kuripan West Nusa Tenggara province of Indonesia, is a development area pockets buffaloes potential, with the support of the agricultural land is quite wide, available forage and waste from agricultural products, so as to support adequacy as fodder for buffaloes. Problems see above it, the purpose of this paper is to determine the estimated value of the selling price of live cattle buffalo, weight and price of farmers, the income economic analysis, which is a farmers' skills in assessing the buffalo live cattle, which need to be understood buffalo by livestock farmers.

## Materials and Methods

### Buffalo Livestock Business Analysis Method

To measure the parameters of the cost structure of livestock farming buffalo breeders, calculated based on the scale of business, so it can be in predication how much profit is obtained by farmers, the income can be defined as the difference between revenues and total costs have been incurred, referring to the data obtained by field survey results, which supported the research Chilonda *et al.* (2001) and Squires (2003).

As for measuring the value of the sale price buffaloes live, in economic feasibility used several factors that can estimate the weight of a buffalo and buffalo prices with a simple formula: Agus (1998),

Weight =  $0.015 \times Pt \times Ld$  and Price =  $0.45 \times Bk \times HDK$ , Remarks:

Pt = body length (cm)

Bk = weight (kg)

Ld = chest circumference (cm)

Hdk = price of meat (Rp)

The percentage values were measured by the individual sex to total number of animals. This count provides an easy way to assess the value of the sale price buffaloes live, so as to facilitate the sale and purchase of buffaloes, between consumers and producer the ultimate goal is to get the optimum benefit both consumers and producers.

## Results and Discussion

### Farmers Characteristics

Farmers in the District of West Lombok Kuripan have diverse characteristics, in terms of age, level of formal education passed by, Sd, number of family members, status and extensive tenure and experience farm animals, age of farmers is different in the age group productive average about 45 years. The condition is one of the aspects that support livestock farming activities. Rusdiana *et al.* (2012), argues that, on average farmer age of was still production and has the potential to do other livestock farming activities.

### Buffalo The Average Ownership

This situation illustrates the pattern of the parent buffalo cattle raising efforts, to get their of spring, while buffalo cattle sold on a young male enlargement to the great days ahead as Idul Adha and the New year shown in Table 1.

Table 1. The average farmer's ownership of buffaloes (n-23)

Description	Number of animal (head)	Mean (head)	Percentage (%)
Adult female	42	1.82	62.67
Young female	10	0.43	14.92
Female child	2	0.09	2.98
Adult male	7	0.30	10.45
Young male	4	0.17	5.97
Male child	2	0,09	2.98
Total	67	2.9	100

Table 1. the average ownership Kuripan buffalo in the Distric of west Lombok Regency of the population that maintained the highest of visible stem tail approximately 1.82 or (62.67%) ou farmers as his successor 0.43 tail (14.92%), male around the 0.30 (10.45%). There are some farmers and is one person who is not heavy, parent livestock reared never given birth or who pregnant less produce for seen in farmers Table 2.

Table 2. Response or the response of farmer to venture

No	Description	Number of respondents (n-23)	Percentage (%)
1.	Likes to keep	6	26.09
2.	Very pleased to maintain	14	60.87
3.	Ordinary	2	8.69
4.	Not happy	1	4.34
Total			100

Table 2. indicates that buffalo cattle farmers response to the responses and opinions of farmers in an aging the business of raising livestock buffalo, it appears that the states heavy about (20.09%) and very heavy (60.87%).

### Buffalo of Feasibility

The results of this economic analysis of facilitate how to calculate the numerical value price buffaloes, sales can through middlemen, market directly to slaughter houses (RPH) with the count, heavy =  $0.015 \times \text{Pt} \times \text{L}$  and prices =  $0.45 \times \text{Bk} \times \text{Hdk}$ , Table 3.

Table 3. Mean estimated body weight of buffalo and selling price in farmer's level

Variables	Head (n-23)	Weight (kg)	Pt (cm)	Ld (cm)	Bk (kg)	Live Price (Rp.000)	Hdk (Rp.)	Total Price (Rp.000)
Adult female	42	0.015	131.4	191.3	377.05	0.45	89.500	15.19
Young females	10	0.015	126.2	176.3	333.74	0.45	89.500	13.44
Female chid	2	0.015	-	-	78,7	0.45	89.500	3.17
Adult male	7	0.015	142.4	198.7	42442	0.45	89.500	17.09
Young males	4	0.015	130.3	187.6	366.66	0.45	89.500	14.67
Male chid	2	0.015	-	-	87.5	0.45	89.500	3.52
Total	67	-	-	-	-	-	-	-

Pt= body lengt, Ld= chest size, Bk= bodyweight, Hdk=price of meat

Table 3, shows that the highest value on the sale of livestock buffalo cattle around Rp. 17.09 million, the value of the price of meat around Rp. 89.500,- while for the measurement of buffalo children are not calculated value of body length and chest girth since, has not been able to production used as meat, can be seen with the body weight of cattle. Amik *et al.*, (2006) states that, by using the method of economic analysis is comparative figures between the value of the scale of business or costs incurred in a business for one year.

## Conclusion

The results of the buffalo livestock enterprises by assessing the sales value and the value of body weight was indirectly highly profitable to farmers and also as a booster to maintain the existence of buffaloes. The highest sale value was on male buffalo i.e. Rp. 17.09 million, and the current meat prices was around Rp. 89.500,-. As for the calves were not measured body length and chest girth because it can not be used for meat production, and only could be seen on body weight.

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# Women, Gender Equality in Livestock Development: Case Study from Papua and Central Java

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## Abstract

*Livestock and its products accounted for 12 percent of the agricultural gross domestic products. The number of male and female workers increased by 2.19% and 3.54% per year respectively within the period of 2007-2011. Livestock is an entry point for promoting gender equality and women empowerment in rural areas. The objectives of this study were to analyze the function of livestock for men and women, various roles played by gender in livestock management, and gender issues, including women's empowerment in livestock development. Rapid rural appraisal method was used to collect information from farmers including gender participants in Bintuni Bay, West Papua and Klaten, Central Java. The results revealed that large animals were owned and managed by men, while small animals (goats, sheep and poultry) were mostly kept by women near the household. Women played significant contribution in management, processing and marketing of animals and products. Most of the decision making was by women, while men participated in coordinating activities related to large ruminants husbandry. More than 40% of women had access to economic resources, and often played significant roles in family income. The main constraint's prohibited women to actively involved as leaders in livestock organization were mainly due to social and cultural reasons. Participation of women in livestock activities could increase household income by 10 to 15%. Involved women in livestock sectors were essential, taking into account the level of knowledge there experiences, including specific trainings in livestock marketing and supply chain.*

*Keywords: gender equality, livestock development*

## Introduction

Government policy for livestock development in the framework of national food self-sufficiency are directed to the fulfillment of livestock-based food through community farming programs. Livestock commodities kept for food and the development of agricultural farming were included cattle, buffaloes, goats, and poultry. In the meantime, commodities for export were goats, sheep, pigs, and poultry (Director General of Animal Husbandry and Health or DGAH, 2014). In Indonesia, from 2008 to 2012, the livestock sub-sector and the results contributed to Gross Domestic Product (GDP) around 12% to the overall agriculture (Ministry of Agriculture, 2012).

Livestock enterprise was one of the main activities in rural areas, more villagers involved in this sub-sector along with the increased GDP. The members of male and female workers increased in the period 2007-2011, 2.19% and 3.54% per year respectively, showing that the number of females working in livestock sub-sector was higher than males. However, the employment in the livestock sub-sector in 2012 was dominated by males workers (56-58%) compared to females (42- 44%). (DGAH, 2013). Livestock business and development were the entry point to promote gender equality and the empowerment of women in rural areas. Traditionally, the division of roles and responsibilities in the livestock business was as follows: males for large livestock (cattle, buffaloes, goats) and females for small livestock (poultry). Community participation was still seen in the narrow context, and even there was still gender bias in the development, considering females to be inferior. Women's participation in the agricultural development was not obvious and they seem to be in a position of being unable to develop their business (Sumarti, 2012). Hence, it is important for all development sectors to implement Presidential Instruction No. 9 of 2000 on Gender Mainstreaming (PUG) in development. This paper aimed to analyze: (1) the function of livestock for men and women; (2) a variety of gender roles in the livestock business; (3) gender issues occurring in livestock development programs; and (4) efforts to empower women in the livestock business.

## Methods

The study was conducted in two different places: (1) Teluk Bintuni Regency in West Papua, specifically in Bintuni District (the kampongs of Iguriji, Gaya Baru, Argo Sigemeray), and in Manimeri District (the kampongs of Atibo Pasamai, Banjar Ausoy); and (2) Klaten Regency in Central Java, specifically Jambakan village in Bayat District and Glagah village in Jatinom District. A qualitative approach was used, supported by quantitative data. The approach was carried out through the community case study method, using data collection techniques, in-depth interviews and group discussions on 5-15 household participants per kampong, adopted from Qoriah and Sumarti, (2008). In Teluk Bintuni Regency, the quantitative data was obtained from 47 respondents who raised more than one type of livestock.

## Results and Discussion

### The Roles of Livestock

Livestock was still secondary part of farmers activities, indicated by the numbers and composition of livestock owned by households in the five kampongs of TelukBintuni, and two villages of Klaten Regency (Table 1).

Table 1. The numbers and composition of livestock in each village

Kampong/ Village	Livestock species	Average no of livestock	Age Structure of native chicken
Teluk Bintuni Regency			
Iguriji	Native chicken,	12	12% chicks, 7% young chickens, 81% hens and cocks
	pigs	3	
Gaya Baru	Native chicken,	34	27% chicks, 35% young chickens, 38 % hens and cocks
	pig	7	
Argo Sigemeray	Native chicken,	15	58% chicks, 5% young chickens, 37% hens and cocks
	goats	4	
Atibo Pasamay	Native chicken,	31	50% chicks, 20% young chickens and 30% hens and cocks
	pig	3	
Banjar Ausoy	Native chicken,	39	34% chicks, 14% young chickens and, 52% hens and cocks
	duck	32	
	cattle	4	
	goats	7	
Klaten Regency			
Jambakan	Goat	n.a	n.a
Glagah	Dairy cattle	n.a	n.a

The results indicated that livestock kept by farmers in Teluk Bintuni Regency quite varied, i.e. native chickens, pigs, goats, cattle, broiler, and duck; while farmers in Klaten raised only goats and dairy cows. The livestock diversity was determined by agro-ecological conditions, culture and the existing breeding programs. The Argo Sigemeray and Banjar Ausoy were transmigration settlement unit areas, and most of them were Javanese community who were Muslim, and used to growing vegetables and rice. The residents of Iguriji, Gaya Baru, and Atibo Pasamay were Papuans, most of them were Christian, and used to planting annual crops and local plants like *kasbi*, *petatas*, taro in their gardens and yards. Jambakan and Glagah village in Central Java were poor areas which received Village Independent Food program. Jambakan was categorized as a dry land, got assistance from Government in goats management, while Glagah as a wetland area, received assistance in goats and dairy cows. Small livestock such as pigs, goats, and chickens were raised for family food security, and most activities were carried out by women, as also reported by Qoriah and Sumarti (2008). However, when the livestock function was increasingly important for the family economic resources, then men will be more responsible. For Papuans, livestock business was still a secondary, part of their daily activities. The main economic activities were still characterized by hunting and gathering, such as collecting nuts, red fruits, and sago, hunting deer, and planting *petatas*, taro and *kasbi* in the fields. Chicken raising, selling live chickens or eggs were mostly done by women, while pig raising was done by men and women for the needs of customary parties where as Lestari and Agusta stated that decision

making for input and management by women. Pigs were sold to fulfill children school needs and Christmas celebrations. For Javanese community, both in transmigration areas and in the two villages in Klaten, dairy cows, and also goats, ducks and chicken were managed by men for family income.

### The Division of Gender Roles in Livestock Business

The roles and responsibilities of men and women in the livestock business in Papuan and Javanese communities can be seen in Table 2.

Table 2. Distribution of Gender Roles in Livestock Activities

Activity	Native Papuan	Java Transmigrant	Java - Klaten
Providing feed and drink for chickens	women	women	n.a
Taking care of chicks and sick birds	women	women and men	n.a
Herding birds out and back into the cage	women	women and men	n.a
Selling chickens and eggs	women	women and men	n.a
Feeding pigs	women	-	-
Selling pigs	women and men	-	-
Finding grass/ herding goats	-	men	women and men
Selling goats	-	men	men
Feeding cattle	-	women and men	women and men
Cleaning the cattle stall	-	men	men
Milking	-	-	women and men
Selling cow milk	-	-	women and men

The roles of Papuan women were mostly on poultry and pigraising, while selling pigs was carried out by both women and men. Javanese transmigrants, women tended to raise chicken together with men, while for cattle and goats, men tended to be more responsible than women. In Klaten, goats were raised by men and women, but men were more responsible in cattle raising. This revealed that livestock as an extra family income was carried out by women, while the responsibility for main economic sources was by men, similar to the results of Fuah (1998) that women were more responsible for small livestock. There was no correlation between household economic status and access to benefits, similar to the report of Yuwono and Prasodjo (2013).

### Gender Issues in Livestock Development Program

Gender equality in livestock development could be seen from the men and women access to and control over resources and benefits of the programs as shown in Table 3.

Table 3. Men and women access to and control over resources and benefits

	Native Papuan	Java Transmigrant	Java -Klaten
Access to resources and benefits			
Land	Adat (communal)	men	men
Input (seed, feed, vaccine)	women	men	men
Training	women	men	men
Farmer group	women	men	men
Market	women	men, women	men
Income	women	women was	women
Control of resources and benefits			
Land	Adat (communal)	men	men
Input (seed, feed, vaccine)	Men	men	men
Training	Men	men	men
Farmer group	Men	men	men
Market	women	equal	men
Income	equal	equal	men

A gender gap was found in the Papuan livestock business, where women had more access to resources and benefits, but the control remains in men hands. Raising chicken was secondary, since the main activities were hunting and gathering. Gender gap also occurred in both Javanese transmigrants and Javanese in Klaten, for different reasons. Women worked on livestock, access to and control over the resources and benefits were more men-dominated. Many Papuans believed that women working only to help their husband to make a living. ILO (2007) stated that gender equality was the enjoyment of equal rights, opportunities and responsibility of men and women, boys and girls in all spheres of life. It was a fairness treatment for men and women, according to their respective needs and interests.

## Conclusion

It can be concluded that: 1) Large animals were kept and managed by men, while goats, and poultry were more woman's domain; 2) Women were responsible in livestock management, processing and marketing; (3) Most work and decision-making of women took place at household level, while men participated in public meetings related to goat husbandry; 4) Women had access to economic resources, and were often important income earners for households. Women faced significant structural and cultural obstacles to becoming effective leaders and gaining access to significant roles in society.

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# The Application of *Tesang* Sharing System at Cattle Farms in Indonesia

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## Abstract

The research aimed to analysis traditional sharing system of the *Tesang* at cattle farms in South Sulawesi Province. This study was conducted from April until June 2014. The population was all farmers in cattle breeding and farmers in cattle fattening who apply *Tesang* system and the sample who apply *Tesang* system over 5 years and amounted to 26 farmers in Pattern 1 at Barru Regency and 24 farmers in pattern 2 at Bone Regency. The results showed that *Tesang* sharing system application in South Sulawesi Province was not contracting, application 2 mechanisms for cattle business and the main motivation *Tesang* system with farmers was increase income for family. Inome from pattern costs higher than pattern parent cows

Keywords: cattle, shareng system, *tesang*

## Introduction

Beef cattle have the greatest contribution as a producer of beef, and during this time have not been able to meet domestic demand which tends to increase every year. Increased demand for beef cattle development opportunities locally with scale agribusiness through a partnership (Suryana 2009; Guolingshi *et al.* 2007). Business development of cattle with a partnership is one alternative to improve the profitability of farmers (Kusnadi 2008) and could overcome some of the problems in the beef cattle business namely small scale cattle raising and limited capital (Kariyasa 2005; Mersyah 2005; Suwandi 2005; Kusnadi 2008). Based on problems existed in the beef cattle business, the partnership program is indispensable. Saptana and Ashari (2007) stated that the partnership is a system of alliances the various agribusiness activities ranging from preproduction, production to marketing Beef cattle business partnership pattern or cooperation has been existing in farmer/ranchers society since the first in South Sulawesi Province, known as *Teseng* applied by means someone provide capital in the form of a cow to be developed by others (farmers) with profit sharing mechanism. Usually there is no bond/ written contract about business cooperation between investors and farmers (Pa'teseng). The ccooperation was orally based upon mutual trust and typically are people who are already well known by investors introduced by relatives. *Teseng* system showed that the value of the trust (confidence) played a very important role (Sirajuddin *et al.* 2013), therefore, exploring the values of local wisdom is a strategic effort in building national character (Wagiran 2012).

## Materials and Methods

The study was conducted in April through June 2014 in Barru and Bone district, province of South Sulawesi which is central production of Bali cattle (local cattle). *Primary* data consisted of characteristic data, the sharing system *Tesang* and secondary data were obtained to describe consisted of data from relevant agencies. A qualitative approach was used obtained to describe the procedures, mechanisms and farmers reasons for applying *Tesang* sharing system in beef cattle business. Samples were be cattle breeders who had applied *Tesang* profit sharing system for 5 -10 years, amounting to 24 people for cow's parent and 26 people for system costs.

## Results and Discussion

### Application procedures of Traditional Sharing System (*Tesang*) In Beef Cattle Business

*Tesang* revenue sharing system carried out by cattle ranchers in the province of South Sulawesi has been handed down and carried out by the local community and has been rooted for generations although in

its development has undergone several adjustments to the method of division that is based on mutual trust plus the customary law that supports the application of the *Tesang* system and make the system for can last for a long time in the life of the local community (Sirajuddin *et al.* 2014). Furthermore Saidari (2009) stated that in the agreement whose legal provisions is applied as customary law it is written that a person who is entitled to a land for some reason could not cultivate it himself, but want to get the results could allow others to conduct farming on the land and the result is divided between them according to a predetermined balance

The main thing to note in the application of Tesang profit share system in South Sulawesi Province was:

1. Between the owners and breeders there must be trust that is built up, for the implementation of this system usually there was no written agreement. All agreements were awakened only on an oral agreement between the owners and breeders. Usually breeders keep having a low income level (poor) so that the owners of capital help them by giving a cow or funding to buy a cow. This is appropriate with opinion of Mosher (1997) that the profit share system is a cooperation which is bound by an agreement of sharing 50%: 50%. This system is mostly done because of poverty and difficulty to get capital which force someone to accept his fate to cultivate the land or raise livestock that is not his own.
2. Before livestock owners is entrusting his cows for breeding to a people, they usually have been considering several aspects such as kinship, breeding experience and the availability of forage to meet the needs of cattle
3. The period of application of the Tesang system was not necessarily or adapted to the circumstances. For example, when farmers suddenly needed cash and Teseng system had been local running for two years then at that time could be said to be a deal for ending the system and calves as the results of cows raised is shared according to the previous agreement.
4. Part of each party will shared equally means that in the first year if the female cattle produced calves, then it would be owned by the proprietor and calves resulted in the second year were given to farmers or vice versa. This is appropriate with statement from Hadikusuma (2001) that the agreement for the results of the farm according to the law of treaties customs revenue sharing system commonly applied for this is 50% for providers of calves and 50% for the keepers.

### **Mechanism for sharing system enterpris was based on *Tesang* imposed by cattle ranchers**

In the Bugis society who inhabits most regions in South Sulawesi Province, the term sharing agreements was commonly referred to as “teseng/tesang”. Although the term is the same for Makassar and *Bugis* ethnics but implementation and agreement form tended to vary due to the different understanding and the practice prevailing in the communities are also different. Sirajuddin *et al.* (2014) opinion the description of the pattern of results share applied by cattle ranchers in the province of South Sulawesi is described as follows:

#### **Pattern I (cow’s breeder)**

In this pattern, the cow was given by the owner to breeders for breeding were adult female’s cows that had been calves. Distribution system for the results was that in the first year. The cow that gave birth produced the calf was to the owner, while, in the second year, calves the breeder accept according to conclusion agreements, that until the fifth year each party obtained two cows or more. This pattern was largely applied as by cattle ranchers that produced good cows.

The results revealed that the cattle owners at the beginning before implementing. The system choose farmers who could maintain the cows or these who were erady as a breeders and agree by could capital owners. Owners provided cash to farmers and expected that the money that had been provided will be used to buy cattle in accordance with the type of cow that was desired by capital owners. Under the condition farmers bought cows good quality and easy to raise, with expectation to produce many calves during the production period

#### **Pattern II (costs based systems)**

In this pattern, the livestock or capital owners gave animals to farmers to be fattened within 6-24 months and then sold them where keeper’s obligation is to bear the entire feed (fodder forage and concentrates), treatment and cages. For examples for the revenue sharing system, if within a certain time the bulls were be sold then the income, was reduced by costs for subsequent maintenance. The profit will be divided between the breeders and owners of capital. Actually, any pattern that was applied to the Tesang revenue sharing system for cattle farmers was not a problem to farmers, because they only accepted cattle given by the owners to be maintained and earned revenue to farmers from profit-sharing system. According to Saidari

(2009) in addition to the agreement, crop is not included in the agreement livestock such as cattle and buffalo only. At this Tsystem, where bulls were reared for fattening or breeding with the maintenance period was 6 months, 1 year to 2 years. In the maintenance of the bulls, that buyers bear the costs for maintenance before sold in Bone regency .Farmers who responsible for maintenance cost would sell their cattle in Barru Regency.

### **Reasons for Cattle Breeders applying Tesang Sharing System**

The reasons of farmers to apply the pattern of *Tesang* results share in South Sulawesi Province are as follows:

1. Farmers did not have the capital to buy a cow
2. Cows belonging to the family
3. Farmers wanted to have the cows themselves
4. The desire to maintain a cow and there were sufficient land
5. Filled the free time for the benefit
6. Increased family income
7. Economic demands of Family
8. Venture capital was not enough
9. It was easier to apply the system for *tesang* results share compared to the results share of government
10. Requested of friends or relatives

It was suggested that social factors and economic factors were the rain of cattle ranchers do applied Tesang system. Of the reasons put forward showed different reasons by cattle ranchers in the province of South Sulawesi and the main reason is to increase the income This is in accordance with the opinion of Isbandi (2004), who stated that in general, farming is a series of activities ranchers who manage factors of production such as land, capital, labor, crops and livestock with the aim of obtaining the maximum benefits to meet the needs of families in line also with the research of Saragih (1997), that there are three types of motivation of farmers/ranchers in Garutto be shepherd of cattle sheep, one of which is to increase revenue.

### **Conclusion**

Tesang was sharing system traditional with application not contracting but trust farmers in cattle business. Tesang mechanism were 2 (two) pattern that cows parent system and cost system. Few reason farmers apply Tesang but main reason was increase income.

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**THEME J.**  
**ANIMAL HEALTH**

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# Cattle Importation and the Trend of FMD Occurrence in Peninsular Malaysia from 2000-2010

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## Abstract

*Foot and mouth disease (FMD) is one of the most contagious diseases of cloven-hoofed animals. The disease is considered as one of the most important trans-boundary animal diseases (TADs) due to its contagious nature that can easily spread and be introduced by global animal movements. In the past decade, Peninsular Malaysia has imported a large number of live cattle from various countries to sustain the local cattle industry and ensure adequate animal protein for local consumption. This study found that the increase of live cattle importation from highly FMD endemic country from 2007 to 2010 is consistent with the increasing number of live cattle imported in Malaysia.*

*Keywords: cattle importation, Foot and Mouth Disease*

## Introduction

Malaysia is one of the largest live animal and animal products' importers in the region. The demand for animal protein increases as the populations expand. Animals originating from FMD free countries such as Australia and New Zealand are expensive, and the price is increasing. Alternatively, Malaysia is importing animals from Southeast Asia. Countries such as Thailand and Myanmar reported consistent high numbers of FMD every year. Therefore, importation from these countries would certainly increase the risk of introducing new infection among the local animal populations. The work of Wongsathapornchai (2007) estimated that at least 1 infected animal will be accepted for importation into the MTM zone per consignment. The aim of this study is to describe the cattle importation trends from year 2000 to 2010 and to highlight its relationship with the local FMD occurrence within the same period.

## Materials and Methods

Data of cattle importation and the frequency of FMD occurrence in Peninsular Malaysia from year 2000 to 2010 were collected. All data were obtained from Department of Veterinary Service and ARAHIS via the SEAFMD website (<http://www.seafmd-rcu.oie.int/index.php>).

Data were analyzed using descriptive analysis via frequency measures and tabulations of live cattle, beef and milk commodity demands within year 2000 to 2010. The graphs were plotted to facilitate comparison with other related study findings from the literature. Those graphs were then compared with the endemic graph which produced from the frequency of FMD occurrence in Peninsular Malaysia also within year 2000 to 2010 to see the regularity of the disease pattern that could be due to the importation activity.

## Results and Discussion

The trend of live animal importation has changed in the past 10 years. In early 2000, more animals were imported from countries such as Australia and New Zealand which were declared free from FMD. Starting 2007 onwards larger proportions of animal importation came from Thailand, part of an FMD endemic region. The average cattle population in Malaysia for the second half of the decade between 2000 and 2010 was 746,367. Australia was the biggest supplier of live cattle between 2000 and 2004 but the supply markedly decreased in 2004 by 45%. Thailand became the major contributor by 2007 because of changes in the import policy allowing more cattle to be brought in (Figure 1).

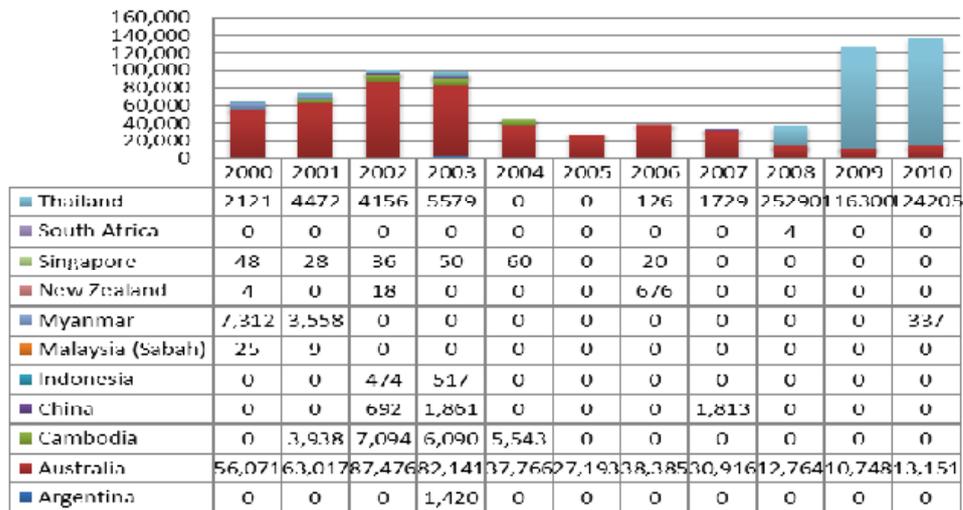


Figure 1. The statistic of live cattle importation into Peninsular Malaysia starting year 2000 to 2010 from various countries (DVS, 2011).

The number of FMD outbreaks in the early 2000 was consistently low but slowly increased following larger proportion of imports coming from FMD-endemic Thailand (Figure 2). The highest FMD occurrences of 137 outbreaks in Peninsular Malaysia in 2008 were recorded and continue to be high in 2009 onwards concurrently with the period when cattle import from Thailand was started to active.

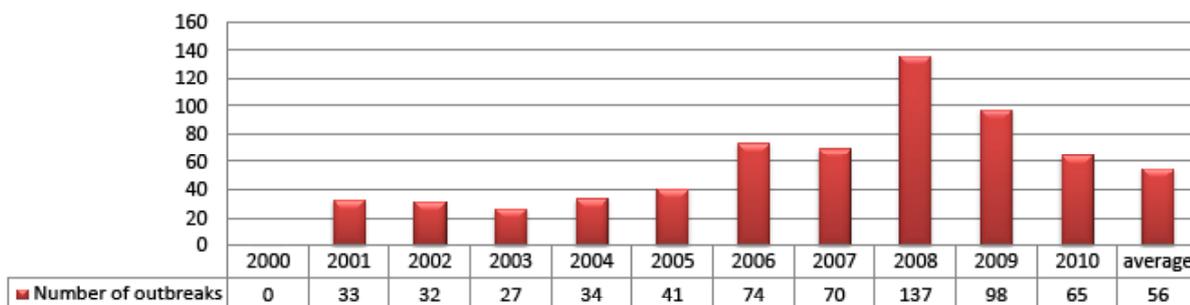


Figure 2. The number of FMD outbreaks in Peninsular Malaysia from year 2000 to 2010 (DVS, 2011)

This study delineates the need to have a better cattle importation protocol and adherence to quarantine protocols if importation from high risk FMD countries were to be continued. As long as the Malaysian ruminant industry remains rudimentary, cattle importation cannot be avoided but measures to reduce the risk of disease introduction can be strengthened to achieve the national goals for specific disease freedom.

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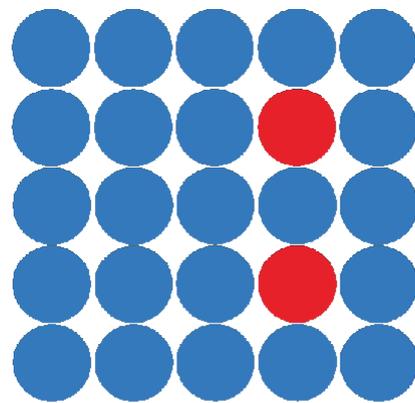
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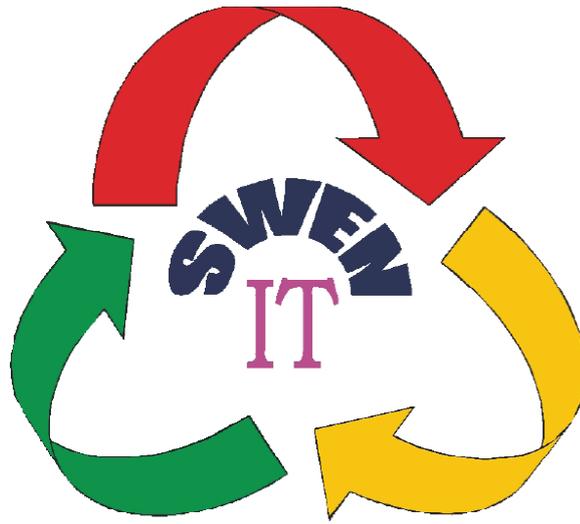




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