

IAEA-TECDOC-1640

# ***Improving Livestock Production Using Indigenous Resources and Conserving the Environment***

*A publication prepared under the framework of a  
Regional Cooperative Agreement for Research, Development and  
Training Related to Nuclear Science and Technology for Asia and  
the Pacific project with technical support of the Joint FAO/IAEA  
Programme of Nuclear Techniques in Food and Agriculture*



**IAEA**

International Atomic Energy Agency

Improving Livestock Production  
Using Indigenous Resources  
and Conserving the Environment



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INTERNATIONAL ATOMIC ENERGY AGENCY  
VIENNA, 2010

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**IMPROVED LIVESTOCK PRODUCTION USING INDIGENOUS RESOURCES  
AND CONSERVING THE ENVIRONMENT**

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## FOREWORD

Livestock farming is very important in Asia and the Pacific region as a source of livelihood for resource poor farmers' — provision of food and food products and as a source of income. However, livestock productivity in many countries is below their genetic potential because of inadequate and imbalanced feeds and feeding, poor reproductive management and animal diseases exacerbated by lack of effective support services, such as animal husbandry extension, artificial insemination (AI) and/or veterinary services.

The International Atomic Energy Agency (IAEA) and the Regional Cooperative Agreement for Research, Development and Training Related to Nuclear Science and Technology for Asia and the Pacific (RCA), with technical support of the Joint FAO/IAEA Programme of Nuclear Techniques in Food and Agriculture, implemented a Technical Cooperation (TC) project entitled “Integrated Approach for Improving Livestock Production using Indigenous Resources and Conserving the Environment” (RAS/5/044). The overall objective of the project was to improve livestock productivity through better nutritional and reproduction strategies while conserving the environment. The specific objectives were (i) to improve animal productivity and decrease emission of selected greenhouse gases, (methane and carbon dioxide) and selected nutrients (nitrogen and phosphorus) into the environment; and (ii) to identify and adopt better breeding strategies that would improve animal productivity.

This publication contains research results presented by scientists during the final review meeting incorporating the contributions of the experts associated with RAS/5/044. It is hoped that this publication will help stimulate further discussion, research and development into ways of improving the efficiency and productivity of livestock thus leading to higher income for smallholder farmers in the region.

The IAEA officers responsible for this publication were O. Perera, A. Schlink and E.N. Odongo of the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture.



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## SUMMARY

### 1. INTRODUCTION

Livestock production is an important source of livelihood for the resource poor farmers in the Asia and the Pacific region. It is a source of food and food products and a source of income. However, livestock productivity in many countries in the region is below their genetic potential, mainly because of inadequate nutrition, poor reproductive management and animal diseases. This is exacerbated by lack of effective support services such as animal husbandry extension, artificial insemination (AI) and/or veterinary services. From a nutritional perspective, productivity of the animals is restricted by imbalanced feed e.g. the low nitrogen (N) and high fibre content of the native grasses and crop residues which is exacerbated by the seasonal availability of the feed resources. Furthermore, low quality, highly fibrous forages when fed to ruminants result in higher enteric methane production than better quality forages, which represents a 5 to 15% loss in gross energy intake depending on type of carbohydrate, addition of dietary fat, quantity of feed ingested, and processing of forages.

Another major limitation to increased milk and meat production in the region is the scarcity of high quality cattle, buffaloes and yaks partly due to the neglect of indigenous breeds i.e. no effort is devoted to improving indigenous breeds through selection for desirable production traits, etc and where cross-breeding has been executed, it is often performed without clearly defined goals such that in some situations the indigenous genetic resource is threatened with extinction. Furthermore, where upgrading of local animals has been undertaken through national cattle and buffalo breeding programmes, there is often a lack of knowledge and/or lack of procedures to ensure optimum use of the improved offspring from such programmes.

### 2. BACKGROUND AND OBJECTIVES

Technologies that can be used to alleviate some of these problems have already been developed through national and/or regional projects. For example, under the project RAS/5/035, "Improving Animal Productivity and Reproductive Efficiency" (1999-2004), urea molasses multi-nutrient block (UMMB) technology was widely disseminated and adopted by farmers in the participating countries. New, lesser-utilized and lesser-known plants capable of growing in poor and degraded soils were identified, evaluated and used as animal feeds. Furthermore, the UMMB was used as a carrier for anti-helminthic agents and for controlling gastrointestinal tract parasitic load to enhance livestock productivity. A computer database application (Artificial Insemination Database Application; AIDA Asia) was also developed for recording, analyzing and interpreting field and laboratory data and was transferred to national AI services.

The current project was focussed on identifying simple strategies that would i) decrease enteric methane and carbon dioxide production from ruminants and channel the energy for increasing milk and meat production as well as improving reproduction, ii) increase N retention in the animal thus lowering N loss into the environment, and redirection of N excretion in a manner that a higher proportion of N goes to faeces than into urine, iii) decrease loss of N and P into the environment from manure through appropriate manure management strategies, and iv) create inventories for methane production from ruminant animals.

## **2.1. Meetings**

The first meeting to plan project activities was hosted by the Institute of Agricultural Environment and Sustainable Development of the Chinese Academy of Agricultural Sciences (CAAS), Beijing, China and was held from 4 to 8 April 2005. It was attended by 23 project counterparts from 12 RCA Member States (Bangladesh, China, India, Indonesia, Myanmar, Mongolia, Pakistan, Philippines, Sri Lanka, Thailand and Vietnam) and was supported by three IAEA experts and two IAEA Technical Officers. Each participating Member State nominated two project co-ordinators, one for animal nutrition and the other for the reproduction/breeding component.

The final project meeting was held in Jakarta, Indonesia from 5 to 9 May 2008 hosted by the National Nuclear Energy Agency of Indonesia (BATAN) to: (a) review results (animal nutrition and reproduction/breeding) obtained in each Member State; (b) assess the achievements, outputs and potential outcomes; (c) draw conclusions and recommendations; (d) list the activities that should be continued using national and other resources; and (e) prepare manuscripts emanating for publication in an Agency TECDOC. The meeting was supported by two IAEA experts (Dr. Karen Marshall of Australia and Prof. Singh Nanda of India) and the two IAEA Technical Officers (A. Schlink for nutrition and O. Perera for reproduction/breeding).

## **2.2. Other activities**

The following group activities were held during the course of the project:

- Experts/Consultants meetings:
  - Manure management, December 2005, Ho Chi Minh City, Vietnam;
  - Selection criteria for breeding heifers, February 2006, Mymensingh, Bangladesh;
- Training courses:
  - Methane production methodologies, April 2006, Khon Kaen, Thailand;
  - Selective breeding and gene technologies, April 2006, Daejeon, Korea.

## **3. SYNTHESIS OF RESULTS**

The topics addressed by participating Member State projects in the animal nutrition and reproduction/breeding components are presented in Tables I and II, respectively. Several countries conducted baseline surveys and most were involved in feed supplementation studies and methane production measurements. A summary of the results and outputs from the nutrition and reproduction/breeding components are presented in Tables III and IV, respectively. The training activities conducted for farmers and agricultural extension personnel in each Member State is presented in Table V.

TABLE I. NUTRITIONAL ISSUES ADDRESSED IN EACH PARTICIPATING MEMBER STATE

Activity	BGD	CPR 1	CPR 2	INS	MO N	MY A	PAK	PHI	SRL	THA
Baseline surveys	X	-	X	-	-	-	X	-	-	X
Feed supplementation	X	X	X	X	X	X	X	X	-	X
Animal production	X	X	-	X	X	X	X	X	-	X
Methane production	X	X	X	X	-	-	X	-	-	X
Manure management	X	X	X	X	-	X	X	X	X	X
Manure quality	X	-	-	-	-	X	X	X	X	X
Soil fertility	-	-	-	-	-	-	X	X	X	X
Forage/crop production	-	-	-	X	-	X	X	X	X	X
Nuclear techniques	-	-	-	X <sup>a</sup>	-	-	X <sup>b</sup>	X <sup>c</sup>	-	-
Economics	X	-	-	X	-	-	X	X	-	-
Training	X	X	X	-	-	X	X	X	X	X
Extension	X	X	X	X	-	X	X	X	X	X

BGD = Bangladesh, CPR1 = China, Hangzhou, CPR2 = China, Lanzhou, INS = Indonesia, MON = Mongolia, MYA = Myanmar, PAK = Pakistan, PHI = Philippines, SRL = Sri Lanka, THA = Thailand  
<sup>a</sup>Rice developed by mutation breeding; <sup>b</sup>Radioimmunoassay; <sup>c</sup>N-15 studies

TABLE II. REPRODUCTION/BREEDING ISSUES ADDRESSED IN EACH PARTICIPATING MEMBER STATE

Activity	BGD	IND	INS	MON	MYA	SRL
Baseline surveys	X	X	X	-	X	-
Participatory approach	X	X	X	-	X	-
Dissemination of superior genotypes	X	X	X	X	X	X
Fertility improvement	X	X	-	X	X	X
Health care improvement	X	X	X	-	X	-
Productivity improvement	X	-	-	X	X	X
Advanced reproductive technologies	X	-	-	-	-	X
Genetic conservation	-	-	-	-	X	X
Radioimmunoassay	-	X	X	-	X	-
Enzyme immunoassay	X	-	-	-	-	-
Databases	X	X	X	-	X	-
Economics	X	-	-	-	-	-
Training	X	X	X	-	X	X
Extension	X	X	X	-	X	X

BGD = Bangladesh, IND = India, INS = Indonesia, MON = Mongolia, MYA = Myanmar, SRL = Sri Lanka

TABLE III: NUTRITION ACTIVITIES AND RESULTS/OUTPTS IN PARTICIPATING MEMBER STATES

Country	Activities	Results/Outputs
Bangladesh	<ul style="list-style-type: none"> <li>- Supplementation of rice straw with medicated UMMB</li> <li>- Low-cost shed and moist soil absorbent for manure</li> </ul>	<ul style="list-style-type: none"> <li>- Increased ADG by 50% with feed:gain ratio of 0.13, and increased milk yield by 23% with feed:gain ratio of 0.75; reduced methane production by 23%</li> <li>- Increased N and P content by 40%.</li> </ul>
China - Hangzhou	<ul style="list-style-type: none"> <li>- Supplementation with tea saponins</li> </ul>	<ul style="list-style-type: none"> <li>- Reduced methane production by 30% and improved ADG by 30%</li> </ul>
China - Lanzhou	<ul style="list-style-type: none"> <li>- Baseline survey on soil pollutants</li> <li>- Supplementation with coconut oil</li> </ul>	<ul style="list-style-type: none"> <li>- Database provided to Government authorities</li> <li>- Reduced methane production by 40-50%</li> </ul>
Indonesia	<ul style="list-style-type: none"> <li>- Multi-nutrient supplement</li> <li>- Covering of manure</li> </ul>	<ul style="list-style-type: none"> <li>- Increased ADG by 25% and improved FCR by 20-60%, reduced methane production by 15-70%</li> <li>- Improved rice yield by 40%</li> </ul>
Myanmar	<ul style="list-style-type: none"> <li>- Supplementation of rice straw with Rumex-K</li> </ul>	<ul style="list-style-type: none"> <li>- Increased ADG by 15%</li> </ul>
Pakistan	<ul style="list-style-type: none"> <li>- Supplementation with concentrates using indigenous ingredients</li> <li>- Covering of manure</li> </ul>	<ul style="list-style-type: none"> <li>- Increased DMD by 8% and feed:gain ratio by 39-49% and decreased methane by 14%.</li> <li>- NPK profile improved in manure and soil and fodder production increased by 26%.</li> </ul>
Philippines	<ul style="list-style-type: none"> <li>- UMMB supplementation</li> <li>- Providing shelter and absorbent for manure</li> </ul>	<ul style="list-style-type: none"> <li>- Increased monthly milk yield by 26% and reduced CI by 86 days.</li> <li>- Increased soil porosity and organic matter and increased corn yield.</li> </ul>
Sri Lanka	<ul style="list-style-type: none"> <li>- Providing shelter and absorbent for manure</li> </ul>	<ul style="list-style-type: none"> <li>- Reduced N, P and K losses by 4, 9 and 63% respectively, and improved fodder dry matter yield by 30%.</li> </ul>
Thailand	<ul style="list-style-type: none"> <li>- Supplementation with cassava hay</li> <li>- Covering manure and making bio-fertilizer</li> </ul>	<ul style="list-style-type: none"> <li>- Decreased methane by 12% with no effect on milk production</li> <li>Improved cassava forage by 9%, cassava tuber by 26% and legume biomass by 12%</li> </ul>

TABLE IV: REPRODUCTION/BREEDING ACTIVITIES AND RESULTS/OUTPUTS IN PARTICIPATING MEMBER STATES

Country	Activities	Results/Outputs
Bangladesh	<ul style="list-style-type: none"> <li>- Baseline survey on factors constraining dairy production</li> <li>- Establishment of community based veterinary service</li> <li>- Economic analysis of farming systems</li> <li>- Dissemination of superior cattle genotypes</li> <li>- Progesterone ELISA to monitor reproductive management of buffalo</li> <li>- Adaptation of in vitro embryo production</li> </ul>	<ul style="list-style-type: none"> <li>- Identified and prioritized factors</li> <li>- Increased farm income by \$ 8 and 24/month per cow and buffalo, respectively.</li> <li>- Net income per year was \$ 47/indigenous cow and \$ 361/buffalo</li> <li>- AI done on 219 cows with 56% CR</li> <li>- Oestrus detection rate by farmers was 30%</li> <li>- Under study</li> </ul>
India	<ul style="list-style-type: none"> <li>- Baseline survey on factors affecting onset of puberty</li> <li>- Overcoming sub-oestrus with Ovsynch protocol</li> <li>- Monensin supplementation during summer season</li> <li>- Pre-partum supplementary feeding with monensin medicated UMMB</li> </ul>	<ul style="list-style-type: none"> <li>- Identified and prioritized 14 factors</li> <li>- CR of 67%</li> <li>- Increase in CR by 30%</li> <li>- Increase in CR by 60%</li> </ul>
Indonesia	<ul style="list-style-type: none"> <li>- Application of AI and database</li> <li>- Tissue and DNA gene-bank for livestock</li> <li>- In vitro embryo production and semen sexing</li> </ul>	<ul style="list-style-type: none"> <li>- Improved S/C from 3.2 to 2.5</li> <li>- Over 3,000 samples of genomic DNA stored</li> <li>- Studies initiated</li> </ul>
Mongolia	<ul style="list-style-type: none"> <li>- Introducing AI to introduce a new cross-bred genotype</li> </ul>	<ul style="list-style-type: none"> <li>- Superior offspring produced (57 to date)</li> </ul>
Myanmar	<ul style="list-style-type: none"> <li>- Survey on factors affecting fertility to AI</li> <li>- Development of selection guidelines for AI bulls and their use</li> </ul>	<ul style="list-style-type: none"> <li>- Identified 4 factors</li> <li>- Guidelines available and 8 bulls selected from 38 born</li> </ul>
Sri Lanka	<ul style="list-style-type: none"> <li>- In vivo embryo production and transfer in goat</li> <li>- Development of semen cryobanking technology for conservation of indigenous animals</li> </ul>	<ul style="list-style-type: none"> <li>- Superior offspring produced (5 to date)</li> <li>- Germ-plasm bank initiated</li> </ul>

TABLE V. TRAINING CONDUCTED FOR FARMERS AND AGRICULTURAL PERSONNEL (NUMBER OF COURSES / NNUMBER OF PARTICIPANTS)

Country/Project	Extension			
	Farmers	workers	Technicians	Professionals
Bangladesh – Nutrition	3/45	1/20	-	-
Bangladesh – Reproduction	1/10	-	1/2	-
China – Nutrition Hangzhou	-	-	-	-
China – Nutrition Lanzhou	2/15	2/6	1/3	-
India – Reproduction	5/30	-	6/30	4/20 <sup>a</sup>
Indonesia – Nutrition	2/15	-	2/2	-
Indonesia – Reproduction	1/30	2/25	2/25	-
Mongolia – Nutrition	-	-	-	-
Mongolia – Reproduction	-	-	-	-
Myanmar – Nutrition	1/8	1/10	2/6	2/10
Myanmar – Reproduction	-	-	-	1/20
Pakistan – Nutrition	3/400	2/70	-	3/75
Philippines – Nutrition	2/15	1/10	-	1/15
Sri Lanka – Nutrition	4/120	2/40	-	-
Sri Lanka – Reproduction	1/40	-	-	5/150
Thailand – Nutrition	3/140	3/18	-	1/18 <sup>b</sup>
<b>Total</b>	<b>28/868</b>	<b>14/199</b>	<b>14/68</b>	<b>17/308</b>

<sup>a</sup>Includes three international staff; <sup>b</sup>RCA regional training course

## 4. FINDINGS AND CONCLUSIONS

### 4.1. Nutrition

#### 4.1.1. Findings

- Feed supplementation and rumen manipulation (e.g. with coconut oil, plant saponins, UMMB, multi-nutrient feed supplement, concentrates, etc) improved nutrient utilization and productivity of the animals and reduced enteric methane production in Bangladesh, China, Indonesia, Pakistan and Thailand.
- Proper manure management, i.e. provision of cover and using an absorbent improved manure quality by improving N, P and K retention which in turn improved crop productivity in Bangladesh, China, Indonesia, Pakistan, Philippines, Sri Lanka and Thailand.
- Application of animal manures in the fields improved soil physical and chemical characteristics and crop response in Indonesia, Pakistan, Philippines and Thailand.
- Herbal extracts of selected indigenous plants reduced internal parasite burden in Mongolia.

#### 4.1.2. Constraint

- Because of the need for multi-disciplinary collaboration and because most projects involved the setting up of new techniques, the project duration was insufficient to complete some of the anticipated work to a stage where concrete recommendations could be formulated.

#### *4.1.3. Further Research*

- In vivo estimation of enteric methane production should be investigated further e.g. the use of feed-based agents or additives (natural and synthetic) on effects on methane production is warranted.
- Additional manure management strategies/options should be developed and transferred to farmers to ensure manure quality and minimized environmental pollution.
- Tested methane mitigation and manure management strategies should be evaluated at the field level including measurement of greenhouse gas (GHG) from manure during storage.
- Further research on effects of herbal extracts on internal parasites and animal productivity should be initiated.

### **4.2. Reproduction**

#### *4.2.1 General conclusions*

- The objectives were quite ambitious and there was insufficient time and resources to achieve them.
- The training on selective breeding and gene technologies held in Daejeon (Korea) could have been improved by focusing on the activities to be conducted during the project.
- The outcome of the consultants' meeting on selection of breeding heifers in Mymensingh (Bangladesh) was to be summarized in to a publication but no specific recommendations have been communicated to the PC.

#### *4.2.2. Country specific findings and conclusions*

Country specific findings and conclusions are presented in Table VI.



TABLE VI. COUNTRY SPECIFIC FINDINGS AND CONCLUSIONS

Country	Findings	Conclusions
<b>Bangladesh</b>	<ul style="list-style-type: none"> <li>- Factors constraining dairy production as an industry were prioritized as (1) lack of vet. services, (2) lack of AI services, (3) lack of training.</li> <li>- Community based vet services increased farmers' income by decreasing calf mortality and increasing milk production.</li> <li>- AI technology successfully demonstrated for production of F1s using imported semen; tracking systems in place to record pedigree.</li> <li>- Significant progress made in the standardization of a protocol for in-vitro production of embryos; however, in-vitro culturing requires further work. The long-term objective is to provide an enabling technology for upgrading and supply of F1s.</li> </ul>	<ul style="list-style-type: none"> <li>- Veterinary service program should be strengthened within the country, via replication of the community-based model developed in this project.</li> <li>- Studies should be conducted to identify and implement appropriate genetic improvement systems (breeding programs), such that problems associated with indiscriminate cross breeding are mitigated.</li> <li>- In-vitro production of embryos is not yet mature enough to be applied, and efforts to standardize the technique should continue. At a later stage, an economic analysis should be performed to determine where it is cost beneficial to apply the ET technology.</li> </ul>
<b>Indonesia</b>	<ul style="list-style-type: none"> <li>- Ovsynch protocol is successful in increasing fertility in sub-oestrus buffaloes in a cost-effective manner.</li> <li>- Supplementary feeding with monensin enhanced the efficiency of the Ovsynch protocol and improved onset of post-partum fertility.</li> <li>- Delayed onset of puberty in buffaloes is a serious issue; calf-hood disease was identified as a major causative factor.</li> </ul>	<ul style="list-style-type: none"> <li>- Strategies should be developed to extend the Ovsynch technologies to the smallholders, as these represent the majority of buffalo keepers.</li> <li>- Strategies should be developed to promote supplementation with monensin (especially during the peri-partum period), to promote proper vaccination against calf-hood diseases and appropriate management.</li> </ul>
<b>Indonesia</b>	<ul style="list-style-type: none"> <li>- The use of a database to improve AI recording and management improved services per conception rate from 3.2 to 2.5.</li> <li>- A tissue and DNA gene-bank for livestock has been initiated. Studies on in vitro embryo production and semen sexing have been initiated.</li> </ul>	<ul style="list-style-type: none"> <li>- Record keeping on AI should be promoted to enhance reproductive performance.</li> <li>- Government should provide sustained support to the gene-bank initiative.</li> </ul>

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<b>Mongolia</b>	<ul style="list-style-type: none"> <li>- The Selenge cattle (synthetic breed) are being crossed with Simmental.</li> <li>- Conception rate to AI was 57% and 56/57 animals gave birth with 4 being assisted. Thus, Selenge can be effectively crossed with Simmental.</li> </ul>	<ul style="list-style-type: none"> <li>- Further characterization of the new genotype (F1) is required to establish its net merit in comparison to the local Selenge. Selenge cows selected for crossing with Simmental must have high BCS and should be pluriparous to avoid calving difficulties. Systems analysis should be performed to determine if a continual F1 crossing system is viable in Mongolia.</li> </ul>
<b>Myanmar</b>	<ul style="list-style-type: none"> <li>- The intensity of vulva swelling, redness and mucus discharge at oestrus were found to be the strongest determinants of conception rate after AI. High vaginal temperature (40.6 – 41.5°C) correlated with the highest conception rates.</li> <li>- Procedures for selection of dairy breeding bulls are in place, based primarily on physical characteristics, growth and semen quality. This is followed by progeny testing for milk production.</li> </ul>	<ul style="list-style-type: none"> <li>- Information on secondary signs of oestrus and timing of AI should be extended to stake-holders such as producers, AI technicians and extension workers.</li> <li>- Within the sire selection scheme, the number of bulls subjected to progeny testing should be increased. Breeding objectives should be defined from approaches such as participatory rural appraisals via matrix rating, and used to help inform the sire selection scheme.</li> </ul>
<b>Sri Lanka</b>	<ul style="list-style-type: none"> <li>- Basic facilities for embryo production and transfer have been established and MOET protocols for goat are being developed, although success rate is currently low. The broader objective is to increase rates of genetic gain via within breed selection.</li> <li>- Semen collection from indigenous species using electro-ejaculation of live bulls or from the epididymis has been achieved. The post-thaw motility of the semen was acceptable (&gt;60%). A germ-plasm bank has been initiated using the semen collected in the above trials.</li> </ul>	<ul style="list-style-type: none"> <li>- More work to standardize the MOET procedure is required to enhance efficacy and increase cost-effectiveness. An economic analysis should be performed to determine where it is cost beneficial to apply the ET technology.</li> <li>- The frozen semen must be subjected to fertility test to ensure its potential use for the future. Sampling and associated data collection strategies should be developed for scale-up of the germ-plasma bank.</li> </ul>

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### 4.3. Overall Conclusions

- The project dealt with several aspects that are important for improving livestock production and conserving the environment in the region.
- The involvement of many RCA Member State in the project facilitated greater awareness and information exchange among the stakeholders.
- Flexibility in the project objectives facilitated each participant to address the priorities in their countries, which was acceptable provided they were within the broad framework of the project.
- The project resulted in capacity building for applying new technologies in several Member State.
- A number of planned activities were conducted but not all objectives could be achieved due to the short life span of the project, inadequate financial support from the IAEA and participating institutes, and disjointed technical support and continuity (the project was adversely affected by changes in PC in many Member State as well as a high turnover in IAEA TO).
- The communication between PC and RCA-coordinators was poor in some Member State.
- It would have been beneficial to conduct a mid-term review meeting, as agreed at the planning meeting in China.
- Future projects of this nature should have a longer duration and more inputs, considering the nature of livestock generation intervals and livestock-environment interactions.
- The PC should communicate the outcomes of this project that have practical application for improving livestock production and mitigating environmental pollution to the relevant national authorities and actively promote their adoption in national livestock development programmes.

# UTILIZING INDIGENOUS RESOURCES IN AN INTEGRATED APPROACH TO IMPROVING LIVESTOCK PRODUCTION WHILE CONSERVING THE ENVIRONMENT

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## Abstract

Studies were undertaken to develop strategies to mitigate methane production from ruminants by improving fermentative digestibility of feeds and manure management to help farmers increase livestock productivity as well as income from livestock and crops. A participatory rural appraisal survey carried out in the study area found that intensive rice production was practiced leading to an acute shortage of green forage and hence rice straw was being fed as the principal roughage source for ruminants. The farmers did not follow any improved system of manure management and heaped all the collected manure in open area. In an on-station experiment, feed intake (total, per cent of live weight (LW) or straw dry matter (DM)) of the herbal fortified urea molasses block (UMB) supplemented group did not differ ( $P > 0.05$ ) from that of the control group. Digestibility of OM, CP and CF were higher ( $P < 0.05$ ) than for the control group. Live weight gain of cattle fed herbal fortified UMB was higher ( $P < 0.01$ ) than that of the control group. Rumen ammonia and VFA concentrations were higher ( $P < 0.05$ ) with the feeding of herbal fortified UMB to cattle. Methane production per kg of digestible DM in the UMB supplemented group was lower ( $P < 0.05$ ) than that of the control group. Improved systems for manure management increased ( $P < 0.05$ ) N, P and K retention in manure compared to the traditional system. Lactating cows fed a straw-based diet supplemented with herbal fortified UMB and legume hay did not alter total DM intake, DM intake per cent LW or straw intake compared to the control group. The increased milk yield of the supplemented group was higher ( $P < 0.01$ ) than for the un-supplemented control group, however, milk composition remained unaffected. The on-farm study with milking cows also showed increased ( $P < 0.05$ ) ammonia N and VFA concentrations and reduced ( $P < 0.05$ ) methane production in the rumen from feeding herbal fortified UMB plus legume hay compared to control feeding. It is concluded that improved feeding increased growth rate and milk yield and reduced ruminal methane production in cattle.

## 1. INTRODUCTION

Global warming and depletion of the ozone layer because of the increased GHG emission is a major concern for the world's human population. Methane is an important component of GHG in the atmosphere, and a significant proportion is produced by anaerobic decomposition of organic matter. Livestock production contributes to the accumulation of methane through fermentative digestion of the rumen, and indirectly when faecal material decomposes anaerobically. Methane production from ruminants is mainly influenced by feed intake and digestibility. Ruminants on low quality feeds possibly produce over 75% of the methane from the world's ruminant contribution to atmospheric methane. To stabilize methane concentration in the atmosphere, it is predicted that methane production needs to be decreased by 10-20%. Therefore, reducing

methane loss during rumen fermentation of fibrous feeds will not only increase the efficiency of utilization of energy but also will reduce environmental pollution.

In Bangladesh there are 23.4 million cattle, 0.82 million buffaloes, 33.5 million goats, 1.11 million sheep, 138.2 million chicken and 13.0 million ducks [1]. Ruminants' account for 28% of the total livestock population and the majority are indigenous breeds with low productivity. This productivity is further reduced because of the severe shortage of green forages with rice straw become the major roughage source for ruminants resulting in higher methane production during rumen fermentation than would occur on green forage-based diets. Despite this situation, little attention has been directed towards rumen methane production, its quantification or reduction in ruminant animals. A recent report [2] found that 82% of the CH<sub>4</sub> production in Bangladesh came from rice and livestock production with 43 percent being due to enteric fermentation and manure breakdown from livestock production. Therefore, if the digestibility of fibrous feedstuffs in ruminant animals can be increased then methane production could be reduced and the combined effect would reduce environmental pollution as well as improve the livestock productivity, thus, improving the livelihoods of the livestock farmers.

This experiment investigates an integrated approach to ruminant production using supplementation and manure management to improve productivity while reducing methane production.

## 2. MATERIALS AND METHODS

### 2.1 Assessment of livestock feeding and management, manure management and socioeconomic conditions of farmers

#### 2.1.1 Selection of area and selection of farmers

Participatory Rural Appraisal (PRA) technique was used in a village called Dattapara (Latitude 23° 43'N Longitude 89° 34'E) in Muktagacha Upazila (sub-district) of Mymensingh district. Two groups of farmers were selected for the PRA study: small farmers and medium farmers. Ten farmers from each of the groups (in total 20) participated in the PRA sessions.

#### 2.1.2 Conduction of PRA

PRA was undertaken to determine landholdings, cropping systems, livestock rearing, livestock feeds and feeding systems, storage of livestock manures and utilization, etc. along with the socioeconomic condition of farmers. The PRA was completed in two sessions: livestock rearing and manure management. The PRA tools used in the study were focus group discussion (FGD), semi-structured interview (SSI) and scored causal diagram.

## 2.2 Effect of improved feeding on intake, digestibility, growth rate and methane production in growing cattle and manure management on manure quality on-station

### 2.2.1 Selection and grouping of animals

Ten growing castrate cattle of similar live weight (120 kg) were selected for a growth and digestibility experiment. The animals were randomly divided into two groups with 5 animals in each group.

### 2.2.2. Preparation of herbal fortified UMB

Leaves of the herbal anthelmintics (pineapple) were collected, washed clean, freeze-dried and then ground. The required amount (200 mg/kg LW of cows) was then added to the ingredients of UMB and mixed thoroughly. The herbal fortified UMB was then prepared using the cold press method [3]. Composition of the blocks is as follows (Table I):

TABLE I. THE COMPOSITION OF COLD PRESSED HERBAL FORTIFIED UREA MOLASSES BLOCKS

Ingredient	Amount (%)
Molasses	39
Wheat bran	20
Rice polish	20
Urea	10
Common salt	5
Lime	6

### 2.2.3 Feeds and feeding of animals

The basal diet for both groups in the growth experiment was rice straw supplemented with 250 g wheat bran, 250 g til oil cake and 2 kg per day of fresh basis of green Dal grass (*Hymenachne amplexicaulis*, 9% crude protein). The nutrient composition of feed ingredients used in the diets is presented in Table II. The cattle fed solely on the basal diet were the control group. The second group was supplemented with 0.5 kg of pineapple fortified UMB per day. The groups were randomly allocated to the diets and the daily dietary allowance was divided into halves with one half being feed in the morning and the second in the afternoon.

TABLE II. NUTRIENT COMPOSITION OF FEED INGREDIENTS USED IN THE DIETS

Feed	Nutrient composition (g/kg DM)					
	Dry matter	Crude prot.	Crude fibre	Ether extract	Ash	Org. matter
Rice straw	89.0	3.5	35.8	2.0	16.1	83.9
Dal grass	20.0	9.0	26.8	2.5	15.4	84.6
Wheat bran	90.2	15.6	8.2	4.8	5.5	94.5
Til oil cake	90.5	34.5	8.5	8.7	12.8	87.2

#### 2.2.4 *Manure management*

Manure from the animals of the two groups was separately collected daily and well mixed before being divided into two portions for storage. One portion was stored under improved managements (pit under a shed with moist soil in between layers of manure) and the other portion stored using traditional methods (open pit without shed).

#### 2.2.5 *Measurements*

The animals were weighed at the beginning of the experiment and weekly thereafter to determine daily live weight gain. Daily intake was calculated from weekly feed intake. Digestibility of feeds was determined by collection and analyses of feeds and faeces from the cattle. Volatile fatty acids (VFA) and ammonia concentrations in the rumen was determined using Gas Liquid Chromatography and Markham distillation, respectively and methane production was measured *in vitro* using the glass syringe method [4]. Nitrogen, P and K analyses of manure were measured using Kjeldahl for N, colourimetric method using spectronic 21 for P and atomic absorption spectrophotometer for K.

### **2.3 Effect of improved feeding on feed intake, milk yield and methane production in cows and manure management on nutrient content of manure under farm conditions**

#### 2.3.1 *Selection and grouping of animals*

Six crossbred (local × Holstein) lactating cows from a farm household in a village were used in this experiment. The average live weight of the animals was 451 kg. They were randomly allocated according to milk yield to two groups of three cows per group.

#### 2.3.2 *Feeds and feeding of animals*

Two diets were formulated using rice straw as basal ingredient with Diet 1 (control) containing as well wheat bran, rice polish, til oil cake, boiled broken rice and Napier grass (*Pennisetum purpureum*). Diet two consisted rice straw with herbal fortified UMB (prepared and fed as in the previous experiment) and khesari (*Lathyrus sativus*) hay with reduced quantities of wheat bran, rice polish and til oil cake and boiled broken rice. The groups were randomly allocated to the diets and the daily diet allowance was divided into halves with one half being fed in the morning and the second half in the afternoon.

#### 2.3.3 *Manure management*

Manure of the animals from the two groups were separately collected daily and well mixed before being divided into two portions for storage. One portion was stored with improved managements (pit under shed with soil between layers of manure) and the other portion in the traditional way (open pit without shed).

### 2.3.4 Measurements

Daily intake of feed was calculated from weekly feed intake data. Milk yield for each cow was recorded daily. VFA and ammonia concentrations in the rumen as well as methane production were measured from cannulated animals reared under on-station condition using the same feeding procedures as described above. Nitrogen, P and K analyses of manure collected from the on-farm experimental cows were carried out as previously described.

The economic evaluation undertaken using market values feed constituents, labour costs and the value of the milk for each treatment group on a per head basis.

## 3. RESULTS

### 3.1 Assessment on the livestock feeding and management, manure management and socioeconomic conditions of farmers

The results of the study are presented in Tables III, IV and V, and Figure 1 and 2. The study found that intensive rice crop production was practiced and therefore, livestock feed production, particularly green grass production was lacking and in short supply. Sixty-four percent of farmers said that lack of improved breeds was a major problem in livestock rearing and 51% said shortage of green grass was a major problem in rearing livestock. Table III shows the root causes of problems in livestock rearing and suggests possible solutions for them. The main cause of the shortage in green grass as well as lack of grazing land is lack of available land due to intensive cultivation of rice for human food production. A possible solution of the problem is to practice integrated rice/forage production. The solution to poor quality feed and fluctuations in feed supply is the cultivation of legume forages and feed storage, respectively. The main cause of lack of scientific feeding to animals is that the farmers have little access to developed technologies and the solution is to transfer developed technologies to rural farmers.

Figure 2 shows the scored causal diagram for the improved management of manures on farms. The root causes for not following improved manure management practices is the lack of knowledge, unwillingness to adopt improved manure management practices and to some extent, financial constraints. The possible solution for these problems is short training courses for farmers to appreciate the importance of improved manure management. The farmers should be motivated by results demonstrated with the improved technologies and financial problem should be overcome by the building of a low cost shed over the manure pit so that rainwater cannot enter into the pit.

TABLE III: PERCENTAGE DISTRIBUTION OF PROBLEMS IN LIVESTOCK REARING REPORTED BY FARMERS

Problem	Small farmers	Medium farmers	All farmers
	%		
Shortage of good quality feed	13	7	10
Shortage of green grass	56	47	51
Lack of improved breeds	66	62	64
Lack of capital	66	62	64



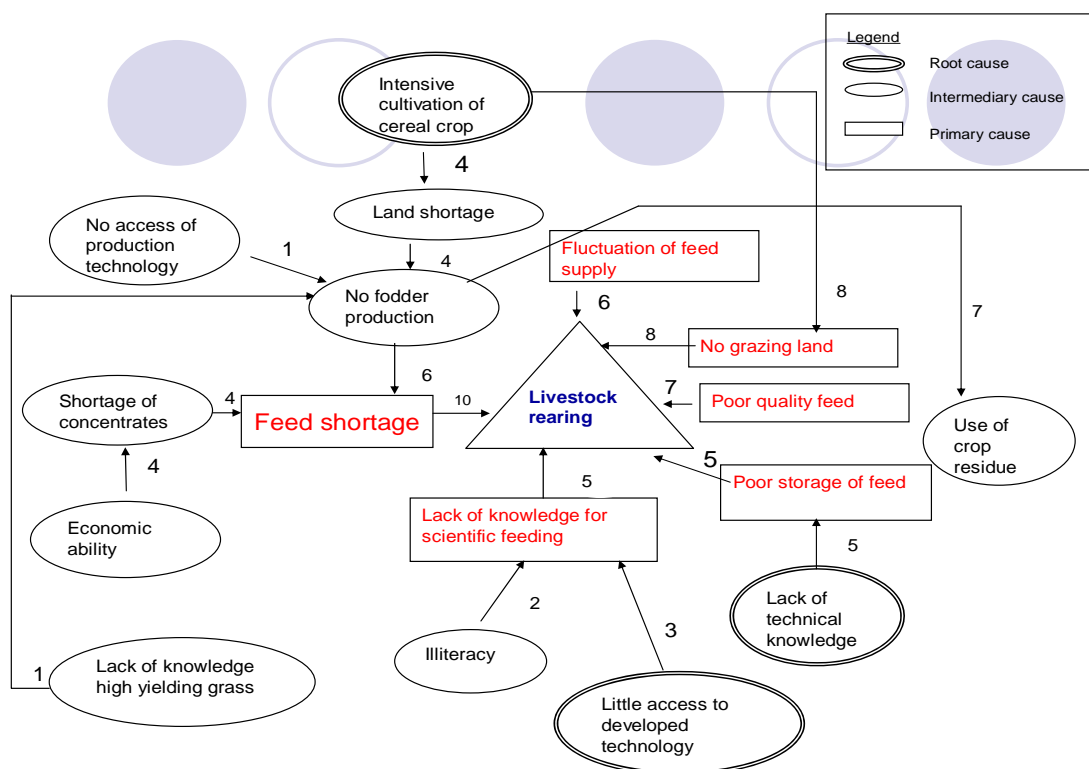


FIG. 1. Scored causal diagram of the problems associated with livestock rearing

TABLE IV: ROOT CAUSE OF PROBLEMS IN LIVESTOCK REARING AND THEIR SOLUTION

Problem	Rank	Score	Root cause	Solution
Feed shortage	1	10	Intensive cultivation of rice	● Integration of forages to rice production
No grazing land	2	8	Same as above	● Same as above
Poor quality feed	3	7	-	● Cultivation of legume forage
Fluctuation feed supply	4	6	-	● Feed storage and conservation
Lack of knowledge of scientific feeding	5	5	Little access to developed technology	● Technology and information transfer among the rural farmers
Storage of feed	6	5	Lack of technical knowledge	● Demonstration of technology at village level.

TABLE V: ROOT CAUSES OF NOT FOLLOWING IMPROVED STORAGE PRACTICES FOR MANURE AND THEIR SOLUTION

Rank	Score	Root cause	Solution
1	5	Lack of knowledge	● Short training should
2	3	Unwillingness	● Motivation should be done through demonstrating result
3	2	Financial involvement	● Low cost shed should be built

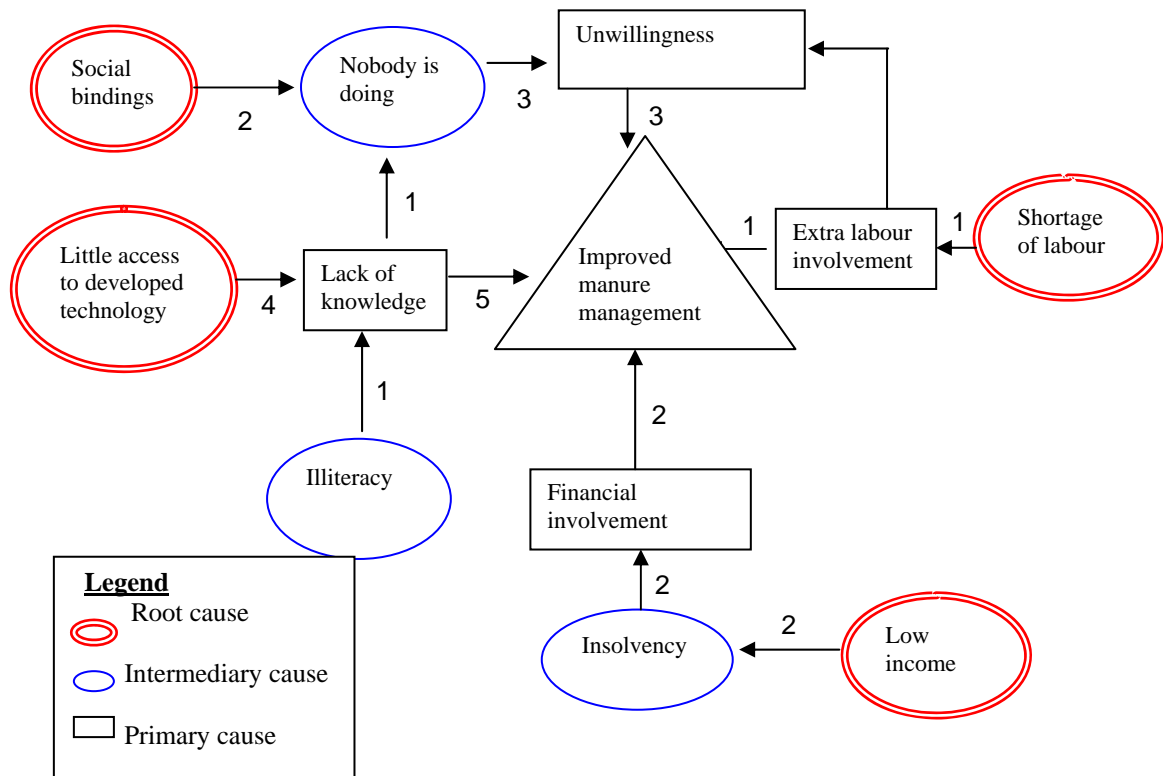


FIG. 2. Scored causal diagram for improved manure management on farms

### 3.2 Effect of improved feeding on feed intake, digestibility, growth rate, methane production in growing cattle and manure management on manure quality on-station

Supplemental effects of herbal fortified UMB with straw-based diets for growing cattle on feed intake, digestibility and live weight gain is presented in Table VI. Total dry matter intake (DMI) of the cattle in the herbal UMB group did not differ significantly from that of the control group. Similarly DMI (as per cent live weight) and total DMI of the UMB treated animal group did not differ significantly from that of the control group. Feeding herbal fortified UMB did not have a significant effect on DM digestibility, however, the digestibility of OM, CP and CF was significantly ( $P < 0.05$ ) higher than that of the control group. The live weight gain of animals supplemented with herbal fortified UMB was significantly ( $P < 0.01$ ) higher than that of un-supplemented control group.

TABLE VI: EFFECT OF SUPPLEMENTING STRAW-BASED DIET WITH HERBAL FORTIFIED UMB ON FEED INTAKE, DIGESTIBILITY AND LIVE WEIGHT GAIN OF GROWING CATTLE

Parameters	Animal groups			Significance
	Control	Supplement	SED	
Live weight (kg)	122.3	120.0	9.3	-
Live weight gain (kg/d)	0.305 <sup>a</sup>	0.458 <sup>b</sup>	0.030	**
Total feed intake (total DMI, kg/d)	3.41	3.48	0.71	NS
DMI (kg/100 kg LW)	2.80	2.90	0.11	NS
Rice straw intake (kg/d)	2.70	2.65	0.09	NS
Digestibility (%)				
DM	58.1	59.1	1.3	NS
OM	62.1 <sup>a</sup>	64.9 <sup>b</sup>	1.0	*
CP	65.8 <sup>a</sup>	67.8 <sup>b</sup>	0.8	*
CF	51.4 <sup>a</sup>	56.5 <sup>b</sup>	1.1	*
EE	56.6	57.5	01.0	NS

<sup>a,b</sup> The values in rows with different superscript differed significantly (at \* is  $P < 0.05$  and \*\* is  $P < 0.01$ )

Table VI shows the concentrations of  $\text{NH}_3\text{-N}$  and VFA concentrations in the rumen of the cattle. Both ruminal ammonia and VFA concentrations were significantly ( $P < 0.05$ ) increased with herbal fortified UMB supplementation. However, in case of the individual VFA concentration, acetate and butyrate proportions were not significantly affected by supplementation, however, the propionate production increased significantly due to supplementation with UMB.

TABLE VII: EFFECT OF SUPPLEMENTING STRAW-BASED DIET WITH HERBAL FORTIFIED UMB ON  $\text{NH}_3\text{-N}$ , AND VFA CONCENTRATION AND PROPORTIONS IN THE RUMEN LIQUOR OF GROWING CATTLE

Parameters	Animal groups			Significance
	Control	Supplemented	SED	
$\text{NH}_3\text{-N}$ (mg/100 ml)	18.72 <sup>a</sup>	22.05 <sup>b</sup>	1.02	<0.05
Total VFA (mM/L)	95.61 <sup>a</sup>	100.95 <sup>b</sup>	1.62	<0.05
	Individual VFA (molar proportions)			
Acetate	67.30	65.31	2.52	NS
Propionate	20.40 <sup>a</sup>	27.52 <sup>b</sup>	1.01	<0.05
Butyrate	7.91	8.12	0.61	NS

<sup>a,b</sup> The values in rows with different superscript differed significantly

*In vitro* total gas and methane production from the experimental diets is shown in Table VII. Total amount of gas produced during fermentation of feeds in cattle decreased slightly, but not significantly with supplementation. However, methane production per kg DDM for the UMB supplemented group was significantly lower compared to that of the control group. Similar results were also observed for per cent methane in total gas production.

TABLE VIII: RUMINAL METHANE PRODUCTION IN GROWING CATTLE AS AFFECTED BY SUPPLEMENTATION OF STRAW-BASED DIET WITH HERBAL FORTIFIED UMB *IN VITRO*

Parameters	Animal groups			
	Control	Treated	SED	Significance
Total gas production (L/kg DDM)	109.2 ± 0.8	104.8 ± 0.5	1.67	NS
Methane (L/kg DDM)	33.4 <sup>a</sup> ± 0.57	25.8 <sup>b</sup> ± 0.3	0.7	<0.05
% of methane in total gas produced (L/kg DDM)	31.1 <sup>a</sup> ± 0.7	24.6 <sup>b</sup> ± 0.7	0.9	<0.05

<sup>a,b</sup> The values with different superscript differed significantly

The water and mineral composition of cattle manure is shown in Table VIII. In the improved system of manure management, there was an increase in N, P and K contents compared to those in traditional system of manure management.

TABLE IX: MINERAL COMPOSITION OF CATTLE MANURE OF TRADITIONAL AND IMPROVED MANAGEMENT SYSTEMS

Minerals	Fresh manure	Decomposed manure	
		Traditional management	Improved management
Moisture (%)	25.5 ± 2.1	18.6 ± 1.2	14.2 ± 1.0
Nitrogen (%)	0.51 ± 0.03	0.85 ± 0.10	1.20 ± 0.12
Phosphorus (%)	0.30 ± 0.02	0.50 ± 0.04	0.70 ± 0.08
Potassium (%)	0.70 ± 0.05	1.25 ± 0.85	1.50 ± 0.15

### 3.3 Effect of improved feeding on feed intake, milk yield and methane production in cows and manure management on nutrient content of manures under farm conditions

Effects of supplementing straw-based diet with herbal fortified UMB on feed intake and milk production of lactating cows is shown in Table X. Total dry matter intake (DMI) of the cows on the herbal UMB and legume hay group was not significantly different from that of the control group. Similar results were also observed in case of DMI per cent live weight as well as straw DMI of the UMB plus hay supplemented group and control group. The increase in milk yield of the cows of the supplemented group was significantly ( $P < 0.01$ ) higher than that of the control group. Fat and protein content of milk was not effected by the supplementation strategy.

TABLE X: EFFECT OF SUPPLEMENTATION OF A STRAW-BASED DIET WITH HERBAL FORTIFIED UMB AND LEGUME HAY ON FEED INTAKE AND MILK YIELD OF CROSSED-BRED COWS ON-FARM

Parameters	Animal groups			Significance
	Control	Supplemented	SED	
Live weight (kg)	453.2	448.0	16.6	-
Total feed intake (total DMI, kg/d)	13.60	13.90	1.03	NS
DMI (kg/100 kg LW)	3.00	3.10	0.48	NS
Rice straw intake (kg/d)	3.52	3.50	0.56	NS
<b>Milk yield (kg/d):</b>				
Average initial milk yield	9.20	8.50	1.32	-
Average final milk yield	9.80	10.50	0.98	-
Total increase in milk yield	0.60 <sup>a</sup>	2.00 <sup>b</sup>	0.20	<0.01
<b>Milk composition:</b>				
Milk fat (g/100g)	4.30	4.42	0.23	NS
Milk protein (g/100g)	3.20	3.35	0.18	NS

<sup>a,b</sup> The values in rows with different superscripts differed significantly

Table XI shows the values for NH<sub>3</sub>-N and VFA concentrations in the rumen of fistulated cattle fed the same diets as was provided to lactating cows on the farm. Both ruminal ammonia and VFA concentrations were significantly ( $P < 0.05$ ) increased by feeding herbal fortified UMB plus legume hay to cows. However, in case of VFA concentration, acetate and butyrate proportions were not significantly affected by supplementation, however, proportion of propionate in the rumen increased significantly with supplementation.

TABLE XI: EFFECT OF SUPPLEMENTING STRAW-BASED DIET WITH HERBAL FORTIFIED UMB AND LEGUME HAY ON RUMINAL NH<sub>3</sub>-N, AND VFA PRODUCTION OF RUMEN FISTULATED CATTLE

Parameters	Animal groups			Significance
	Control	Supplemented	SED	
NH <sub>3</sub> -N (mg/100 ml)	23.82 <sup>a</sup>	26.78 <sup>b</sup>	0.98	<0.05
Total VFA (mM/L)	101.1 <sup>a</sup>	105.3 <sup>b</sup>	1.6	<0.05
Individual VFA (molar proportions)				
Acetate	69.1	69.6	1.6	NS
Propionate	24.4 <sup>a</sup>	27.7 <sup>b</sup>	1.0	<0.05
Butyrate	7.68	8.05	0.67	NS

<sup>a,b</sup> The values in rows with different superscripts differed significantly

Total gas and methane production *in vitro* from fistulated cattle fed the same diets as the lactating cows is shown in Table XII. Total amount of gas produced during fermentation of feeds, although decreased slightly by supplementation, did not differ significantly between the groups. Methane production per kg DDM of the UMB plus legume hay group was significantly decreased compared to that of the control group. Similar results were also observed for percent methane in total gas production.

TABLE XII: EFFECT OF SUPPLEMENTATION OF STRAW BASED DIET WITH HERBAL FORTIFIED UMB AND LEGUME HAY ON RUMINAL METHANE PRODUCTION IN CATTLE

Parameters	Animal groups			Significance
	Control	Supplemented	SED	
Total gas production (L/kg DDM)	112.0 ± 1.01	110.3 ± 1.12	1.7	NS
Methane (L/kg DDM)	31.3 <sup>a</sup> ± 0.4	24.2 <sup>b</sup> ± 0.3	0.7	< 0.05
% of methane in total gas produced (L/kg DDM)	28.0 <sup>a</sup> ± 0.8	21.9 <sup>b</sup> ± 0.8	0.9	< 0.05

<sup>a,b</sup> The values in rows with different superscripts differed significantly

The economic return from feeding supplements to lactating cows is shown in Table III.3.4. The feeding of a supplement returned 98 Taka for every Taka spent to improve the ration and this does not take into consideration the possible improved value of manures produced if improved manure management practices were followed in the dairy farm.

TABLE XIII: ECONOMIC EVALUATION OF SUPPLEMENTATION ON MILK PRODUCTION

Parameter	Control	Supplemented	Difference
Rice straw	10.00	10.00	
Napier grass	30.00	30.00	
Khesari hay	-	30.00	
Broken rice	52.00	39.00	
Wheat bran	27.50	27.50	
Rice polish	15.00	15.00	
Til oil cake	17.00	-	
Molasses	-	2.50	
Cost of feed	151.50	144.00	
Cost of UMB	-	8.00	
Cost of labourer*	50.00	50.00	
<b>Total cost</b>	201.50	202.00	0.50
Increased milk yield (average, Kg)	0.60	2.00	1.40
Income from additional milk selling** (Tk.)	21.00	70.00	49.00

\* The labour cost has been taken as half-day as because the labour also works for other activities of the farmer.

\*\* Milk price: Tk. 35.00 per kg

## 4. DISCUSSION

### 4.1 Assessment of livestock feeding and management, manure management and socioeconomic conditions of farmers in the study area

Intensive rice cultivation practiced by the rural farmers has resulted in no spare land being available for green forage production to feeding livestock and therefore there is huge shortage of green forage. This situation has lead farmers to feed their ruminant animals, particularly cattle and buffaloes with rice straw as major roughage source. Similar results were also reported by [4]. On rural farms in Bangladesh there are number of problems faced in rearing livestock that has lead to lower productivity. The most important of these is the lack of improved breeds and a shortage of animal feedstuffs.

Therefore, breed upgrading programmes through artificial insemination and building awareness of farmers to better feeding and cultivation of high yielding grasses and forages must be introduced in order to improve the livestock productivity in the area. The main reason for lack of ration design in feeding cattle is that farmers had little access to developed technologies. Thus, the proposed solution is to transfer developed technologies in feeds and feeding to rural farmers.

The farmers in the study area, as is the case in other areas of the country, store collected cattle manure as heap near the cattle shed. There is no shed to cover the heap, they do not dig any pit to store the manure, and as a result, manures are sometimes leached by rainfall. However, this is the traditional system of manure storage and management on farms. The main reason for not adopting the improved management systems is the lack of knowledge in manure management. The neglect of this valuable resource can be overcome by awareness building on its utilization by counselling, training and demonstrations in the use of low cost technologies.

#### **4.2 Effect of improved feeding on feed intake, digestibility, growth rate and methane production in growing cattle and manure management on nutrient content of manure on-station**

The non-significant effect of feeding a supplement to the growing cattle indicates that feeding herbal fortified UMB did not lower the roughage content of the diet sufficiently enough to allow the cattle to increase intake. The UMB supplied readily available energy and N to rumen microbes for their digestive activities enabling an improvement in rumen function. This is evident by significant increases in the digestibility of OM, crude protein and crude fibre of the supplemented diet. These increases in the protein and fibre digestibility clearly indicate that the readily available energy and N stimulated rumen microbes to increased digestive activities on the feeds. The improved fermentation is also evident from the significant increases in ammonia N and VFA in the rumen. It can also be observed from the digestibility data that the increase in the digestibility of fibre was the highest of all the digestibility's measured indicating that UMB feeding had a significant effect on improving straw digestibility. The increased digestibility of straw fibre was also evident from the significant ( $P < 0.05$ ) reduction in methane production in the rumen (Table VII). This resulted in significant live weight gain increases due to improved feeding with herbal fortified UMB and further confirms the beneficial effects of UMB on the productivity of cattle. It has been demonstrated from previous work in this laboratory that feeding herbal anthelmintics (Pineapple leaves) to cattle results in a reduction in parasite eggs by more than 80% [5]. Thus, the improvement in live weight gain in the UMB group of animals may have also been due to both anthelmintics effects and improvements in rumen fermentation. The improvement in performance of animals feed UMB has also been reported by others [6]. The N, P and K contents of cattle manures managed under traditional and improved conditions clearly demonstrates that if the manure is stored under slightly improved management systems, such as making a pit, a low cost shed over the pit and spreading soil between layers of manure, will better preserve the manure and increase fertilizer capacity of manures for crops.

### **4.3 Effect of improved feeding on feed intake, milk yield and methane production in cows and manure management on the nutrient content of manure under farm condition**

Improved feeding with herbal fortified UMB and legume hay supplemented diet did not increase feed intake indicating that supplementation did not stimulate intake in lactating cows. Although supplementation had no effects on feed intake of the cows, it did significant effect the productivity of the cows as can be seen from the improved milk yields in the supplemented group. Milk yield of cows supplemented with UMB and legume hay increased significantly ( $P < 0.01$ ) compared to that of the cows from the non-supplemented group. This improvement in the productivity of supplemented lactating cows, like in the cattle growth experiment, is predicted to be due to improved feed digestibility, although digestibility was not measured in the lactation experiment. This hypothesis is supported by a significant increase in ammonia and VFA concentrations in the rumen of the supplemented group which is due to increased microbial activity on fibre and proteins of ingested feeds. The increased milk yields of cows supplemented with UMB is consistent with previous reports using this form of supplementation [7]. The significant reduction in methane production from the rumen contents of cannulated cattle fed on the same diet as in lactating cows indicates that the overall feed particularly the fibrous component has been digested considerably more and is consistent with the growth experiment and lead to increased milk yields in the supplemented group. Simple economic evaluation of the supplementation for milk production found that although supplementation with UMB and khesari hay (*Lathyrus sativus*) involved additional cost, the total cost of the supplemented group's diet was similar to the control since there was a reduction in the quantity of broken rice and removal of oil cake from the supplemented diet. However, supplementation resulted in 1.40 kg more milk than that of the non-supplemented diet leading to Tk. 48.50 more profit than cows on the non-supplemented diet.

It may be concluded from the survey that the farmers in rural Bangladesh feed large amount of straw to their animals due to an acute shortage of green forage. This shortage is due to intensive rice cropping and unavailability of land for forage cultivation. The farmers do not follow any improved system of livestock manure management. The current practice of manure management is to heap manure near the animal shed after cleaning the barn, which results in wastage of manure nutrients that can lead to environmental pollution. Feeding a supplement in the form of herbal fortified UMB to growing cattle significantly improves live weight gain, as well as reducing methane production from the rumen. A similar result was also observed for lactating crossbred cows supplemented with herbal fortified UMB and legume hay leading to increased milk yield and reduced methane production. Improved storage and management of cattle manure to reduce nutrient leaching and degradation results in increased fertilizer nutrient status of manure compared to that from traditional storage systems.

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# ASSESSMENT OF ANIMAL PRODUCTIVITY AND METHANE PRODUCTION USING AN ASSOCIATIVE FEEDING STRATEGY

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## Abstract

Methane production from ruminants is a loss of digestible energy thereby reducing animal productivity and is contributing to environmental pollution. In order to develop a beneficial strategy for improving animal productivity while conserving the environment the present study was conducted to evaluate the effect of a concentrate feeding strategy on the animal productivity and rumen methane production. In this experiment two feeding regimes, Diet-1 and Diet-2, were either fodder alone and with a 10% inclusion of concentrates in the forage diet feed as a phased sequence of 45 days of fodder alone and then 45 days of fodder plus concentrate. The diets were fed to four animal groups comprising of 5 animals in each. Throughout the experimental period, a fresh, chopped fodder of similar age (50-65 days age) was offered to the animals. Average dry matter (DM), crude protein (CP), crude fibre (CF), ash and ether extract contents of the fodder were 21, 9.3, 31.7, 10.1 and 3.1 %, respectively. Diet2 included a concentrate containing 88.0, 16.0, 8.1, 10.0, and 12.1% DM, CP, CF, Ash and EE, respectively. A decrease of 8.2 and 39.5% in group A and B with only minor a minor change in group C and D for feed intake was observed when the animals where feed Diet2. Weight gain for the four groups were 133, 422, 111 and 600 g per animal per day on Diet1 and 244, 688, 177 and 888 g per animal per day on Diet2 for groups A, B, C and D, respectively. With supplementation feeding strategy, there was an increase of 45.4, 38.7, 3.7 and 32.4% in weigh gain over fodder alone diet for groups A, B, C and D, respectively. This was associated with an improvement of 49.7, 62.9, 38.5 and 32% in feed to gain ratio for groups A, B, C and D, respectively. The DM digestibility was 22.9, 4.6, 4.7 and 8.4% higher in groups A, B, C and D, respectively when the groups were feed the fodder diets supplemented with concentrates. On fodder alone, the molar concentration of acetate, propionate, butyrate and valerate was 68, 15, 11 and 23% and when concentrates were included 66, 18, 10 and 1.9% for fistulated cattle. Similarly, acetate, propionate, butyrate and valerate were 62, 19, 13 and 3% on Diet1 and 60, 23, 12 and 2.8 on Diet2 for buffaloes. Concentrate supplementation reduced the acetate to propionate ratio in both cattle and buffaloes. Estimated methane production of 350, 300 g methane per animal/day in cattle and 312 and 278g per animal/day in buffalo on Diet1 and Diet2, respectively. It is concluded that methane losses from various classes of livestock could be minimized with integrated approach to nutrition as well as resulting in a better feed to gain ratio.

## 1. INTRODUCTION

Methane production from ruminant livestock is a direct loss of digestible energy (DE) from the diet, as well as an environmental pollutant contributing to the GHG [8, 16, 18]. Methane and carbon dioxide are produced as by-products during the rumen fermentation process. These gases are not utilized by the animals and are exhaled [17, 19]. The amount of methane produced and excreted by cattle, buffalo, sheep and goat depends primarily on the type of animal, and the amount and type of feed consumed [9, 11]. Enteric rumen fermentation relies on bacteria to breakdown the feed to volatile fatty acids (VFA) and other products that the ruminants can absorb to maintain body functions

and growth. Rumen fermentation enables ruminants to digest coarse plant material (lignocellulosics) that non-ruminants, including humans cannot digest. Because of this rumen fermentation, ruminant animals have the highest methane production per unit of intake of the livestock species evaluated for methane production [6]. Enteric methane production from domesticated livestock species contributed 29% of global emissions in the year 2000 (EPA 2001, EPA 2002; <http://www.epa.gov/fedrgstr>). Methane production from enteric fermentation of livestock species in developing countries, principally cattle, sheep, goats and buffalo was responsible for 64% of global emissions in 1990 and is projected to increase to 73% by 2010 (EPA 2002).

Ruminant productivity in most countries is usually below their genetic potential mainly due to inadequate nutrition. In Pakistan, the majority of ruminants are mostly stall fed fresh fodders using a cut and carry system with no or little access to concentrates [20]. The ultimate result of this feeding system is that there is often under-nutrition resulting in low growth rate and delayed maturity of the animals. This situation reduces the economic returns from livestock and reduces the scope for economic development in the community. There is an increase in ruminant livestock populations in the developing world with many opportunities to improve livestock productivity along with improved environmental management through the abatement of methane production from ruminants [6]. Such improvements may play a role in improving the economic outcomes for smallholder farmers and lead to sustainable environmental management for ruminant livestock. An understanding of rumen microbiology and ruminant nutrition will form the basis for measuring of methane production and abatement in ruminant livestock [2, 13, 18].

VFA including acetate, butyrate and propionate are the primary energy substrates for ruminants. They are absorbed across the rumen wall into the blood stream, converted into glucose in the liver and then utilized by the body as an energy source. Methane, on the other hand, is a by-product of the fermentation process and is released to the air principally through the mouth [11]. The other by-product of this fermentation is microbial protein and this protein is the primary source of protein for ruminant livestock, however protein is also sourced from unfermented (i.e. by-pass) protein that is also absorbed from the small intestine [18]. The production of methane represents a loss of digested energy from ruminant livestock and any reduction in methane production has the potential to improved production efficiency through increased availability of digested energy for productive purposes [6, 8]. This paper assesses animal productivity using an integrated approach to nutrition and estimates its consequences on rumen methane production.

## 2. MATERIALS AND METHODS

### 2.1 Experimental Animals

Four groups comprised of 5 castrate male animals where: group A contained Dajal x Friesian Young (75:25); group B contained Dajal x Friesian adults (75:25); group C contained Sahiwal; and group D contained NiliRavi buffalo (*Bubalus bubalis*). Animals were all of a similar age (1-1.5 years) and were selected from a feedlot at Livestock Production Research Institute, Bahadurnagar, Punjab Pakistan, with average weighs of 221, 435, 387 and 358 kg, respectively for groups A, B, C and D. Parameters measured were feed consumption, weight gain and apparent dry matter (DM) digestibility. A parallel study to the intact animal study was undertaken using two mature Buffaloes and two mature Sahiwal cattle with rumen fistulated where the animals were feed according to same protocols as the intact animals. The animals were fed for a total of

90 days divided into two distinct phases of 45 days each. In phase one the animals were fed *Sorghum vulgare* as a chopped, green fodder (Diet1) for 45 days. This was immediately followed by phase two with the animals being continued on the fodder but also supplemented with a concentrate (Diet2) for a further 45 days.

## 2.2 Feeding Strategies

For the duration of phase one and two the animals were fed fresh fodder *ad libitum* and samples of the offered fodder were taken daily to estimate the DM intake per day. During phase two the animals were supplemented with a concentrate (Anmol Wanda) at the rate of one kg of supplement to every 9 kg of fresh fodder. The composition of the concentrate is shown in Table II and contains a 2 % mineral mixture to overcome mineral deficiencies commonly encountered in fodder fed livestock. Feeds issued were weighed at the time of feeding and residues were collected daily with sub-samples of feeds and residues being stored frozen for further analysis. The animals were allowed unrestricted access to water for the duration of the experiment.

## 2.3 Fodder Production and Composition

*Sorghum vulgare* was grown as the fodder source on a heavy loam soil with the first irrigation at 18 to 21 days after sowing and subsequently as required to maintain fodder growth. Fodder quality was maintained for the duration of the feeding experiment by using sequential sowings to ensure that fodder age at harvest was between 50 and 65 days. The seeding rate was 29.7 kg/hectare and agronomic practices were maintained for the different sowing dates to ensure that fodder quality was maintained for the duration of experiment.

## 2.4 Sample collection and analysis

Representative samples from offered fodder, supplement and residues were analyzed for DM, crude protein (CP), crude fibre (CF), ether extract (EE) and ash contents. The DM was determined by oven drying at 90 °C for 24h. Crude protein of the samples was determined using the Kjeldahl method [22]. Ether extract was determined using Soxhlet apparatus in accordance with [22]. Ash content was determined by ashing at 550 °C in a muffle furnace for 16h [22].

The animals were weighed at the start and then fortnightly for the duration of the experiment. Fodder and supplements were weighed at feeding time and residues collected daily with sub samples of feeds and residues being stored frozen until further analysis. Apparent DM digestibility was determined by total collection of feed and faeces on days 5, 25 35, 60, 70 and 80 of the experiment for both intact and fistulated animals. Total feed, residues and faeces were weighed for each animal, sub-sampled and the samples frozen for further analysis. Digestibility was calculated according to the following formulae:

$$\text{Digestibility (\%)} = (\text{Total feed intake} - \text{Faecal output}) * 100 / (\text{Total feed intake})$$

Rumen liquor was collected via rumen fistulae 15 days after start of phase one and two and from those days repeated thrice at 15-day intervals. Rumen liquor samples were collected 3 hours after the morning feed. The rumen samples were stored at -20 °C until they were analysis. VFA content was determined by gas chromatography using internal standards for acetate, propionate, butyrate and valerate. The gas chromatographic

was a Perkin-Elmer model 3920 with flame ionization detection. The VFA were separated using a glass column packed with stationary phase 15% SP-1220/1% H<sub>3</sub>PO<sub>4</sub> on 100/120 Chromosorb WAW. The inner and outer diameter of the column was  $\phi$  2 and 2 mm, respectively with a total column length of 1.83 metres. The chromatograph was set on isothermal programming with the injection block maintained at 150°C, detector at 250°C and column temperature was maintained at 130°C. The carrier gas was N<sub>2</sub> with a flow rate of 25 ml/minute, and hydrogen supplied to the flame jet at 20 psi and air flow to the detector chamber was 50 psi. Methane production was calculated using rumen stoichiometry inventory from the VFA profile following the method by Czerkaski, 1986 (Pergamon Press, Oxford, 1986).

### 3 RESULTS

#### 3.1 Fodder production and composition

The sequence of six plantings of *Sorghum vulgare* achieved a consistent quality of fodder to provide fresh chopped fodder daily to the animals for the duration of the feeding experiment as shown in Table I. The average DM, crude protein, crude fibre, ash and ether extract content of the fodder was 21, 9.3, 31.7, 10.1 and 3.1%, respectively.

TABLE I. PROXIMATE ANALYSIS OF *SORGHUM VULGAR* FODDER (DIET1) FOR DRY MATTER CONTENT (DM), CRUDE PROTEIN (CP), ETC FOR SIX PLANTINGS AND THE AVERAGE VALUES FOR THE HARVEST

Fodder harvest	S1	S2	S3	S4	S5	S6	Average
Feeding period (days)	1-15	16-30	31-45	46-60	61-75	75-90	90-day average
Composition							
DM (%)	21.2	20.76	20.2	22.0	20.8	21.2	21.0
CP (%)	9.13	9.82	9.7	8.9	9.1	9.4	9.3
CF (%)	30.42	31.46	32.1	32.0	31.5	33.1	31.7
ASH (%)	9.58	10.99	10.2	9.9	10.2	10.2	10.1
EE (%)	3.2	3.6	2.9	3.2	3.0	3.2	3.1

The constituents and composition of the concentrate supplement is shown in Table II. The protein is double and ether extract is four times that of the fodder component of the diet.

TABLE II. THE CONSTITUENTS AND COMPOSITION OF SUPPLEMENT USED IN PHASE 2 OF THE EXPERIMENT

Ingredients	% as fed
Cotton seed cake	10
Wheat Bran	4
Maize gluten 30%	25
Maize (crushed)	10
Torea meal	12
Rice polishing	23
Molasses	14
Mineral mixture	2
Composition	
	% DM basis
Dry matter	88.0
Ash	10.0
Crude fibre	8.1
Crude protein	16.0
Ether extract	12.2

### 3.2 Feed Consumption

The total feed consumptions in phases one and two on a DM basis is shown in Table III. Average feed consumption on a DM basis was 7.3, 20.5, 11.5 and 11.2 kg DM/head per day for groups A, B, C and D, respectively for fodder fed animals (Tables III and IV) and 6.7, 12.4, 11.3 and 11.3 kg DM/animal per day for the fodder feed animals supplemented with concentrates. Supplementation resulted in a decrease in intake of 8.2 and 39.5% for group A and B respectively. However, there was a minor increase in intake of 1.7 and 0.7% in group C and D with concentrate supplementation.

### 3.3 Weight Gain

Weight gains for the four animal groups were variable. Average weight gains for the four groups were 133.3, 422, 111 and 600 g/head per day fed fodder alone. Similarly, supplementation with concentrate in the second phase resulted in large variation between groups with average weight gain of 244, 688, 177 and 888 g/head per day in groups A, B, C and D, respectively. Maximum weight was gained in group D (Buffalo calves) followed by group B. Supplementation with concentrates increase live weight gains by 45.4, 38.7, 3.7 and 32.4% over that of the fodder alone strategy for group A, B, C and D, respectively (Table IV). The results showed an additional gain of 75 kg in Buffalo calves with a supplementation feeding strategy over the fodder alone strategy.

TABLE III. PERFORMANCE DATA FROM 90-DAY FEEDING TRIAL ON DIET-1 AND DIET-2

Phase	1				2			
Feeding Regime	Diet1 (Fodder alone)				Diet2 (Fodder and Supplement in 9:1)			
a.	Feed consumption (DMI, kg per group)				Feed consumption (DMI, kg per group)			
Days	1-15	16-30	31-45	1-45	1-15	16-30	31-45	46-90
Group A	533	563	548	1643	501	522	490	1513
Group B	979	1014	983	4618	922	899	971	2791
Group C	871	1014	983	2604	850	840	870	2561
Group D	832	840	863	2535	840	852	861	2553
b.	Weight gain (kg per group)				Weight gain (kg per group)			
Group A	10	10	10	30	15	20	22	55
Group B	20	30	45	95	35	55	65	155
Group C	5	10	10	25	10	15	15	40
Group D	35	50	50	135	60	65	75	200
c.	Feed/gain ratio (kg Feed/kg weight gain)				Feed/gain ratio (kg Feed/kg weight gain)			
Group A	53.2	56.2	54.7	54.7	33.3	26.0	22.2	27.5
Group B	48.9	33.8	21.8	48.60	26.3	16.3	14.9	18.0
Group C	174.1	101.4	98.2	104.15	85.0	56.0	58	64.0
Group D	23.7	16.8	17.2	18.77	14	13.1	11.4	12.7

### 3.4 Feed to Gain Ratio

The amount of feed consumed per kg of live weight gain for the four groups in the two phases is shown in Table III. In phase one when the animals feed the fodder diet the feed to gain ratio of groups A, B, C and D were 54, 48, 104 and 18 kg feed intake per kg

live weight gain. In phase two when the fodder were supplemented with concentrates the feed to gain ratios were 27, 18, 64 and 12.7 kg feed per kg live weight gain for groups A, B, C and D, respectively. Supplementation with concentrates improved the feed to gain ratios by 49.7, 62.9, 38.5 and 32% for groups A, B, C and D, respectively.

The comparison between diets and the effects of supplementation is shown in Table IV. There are large differences between the animal types in response to the supplement provided in phase two of the experiment. Groups A and B had large declines in feed intake while groups C and D only had minor changes in feed intakes. Group D only had a small increase in weight gain compared to the other three groups in response to supplementation. There was a large improvement in the feed to gain ratio with the feeding of concentrates but once again a large range in improvements from 32.0 to 62.9% for Groups D and B, respectively.

TABLE IV. COMPARISON OF FEED CONSUMPTION, WEIGHT GAIN AND FEED TO GAIN RATIO ON DIET1 AND DIET2

Animal Group	Feed Consumption (kg DM/day/animal)			Weight Gain (g/day/animal)			Feed to Gain ratio (kg DM/kg weight gain)		
Group	Diet 1	Diet 2	Change (%)	Diet 1	Diet 2	Change (%)	Diet 1	Diet 2	Change (%)
A	7.3	6.7	-8.2	133	244	+45.4	54.8	27.5	49.7
B	20.5	12.4	-39.5	422	689	+38.7	48.6	18.0	62.9
C	11.6	11.3	+1.7	111	178	+3.7	104.2	64.0	38.5
D	11.3	11.3	+0.7	600	889	+32.4	18.8	12.7	32.0

### 3.6 Apparent Digestibility trial

Apparent DM digestibility (ADDM) values were 56.1, 65.1, 65.3 and 63.7% for groups A, B, C and D on Diet1 (Table V). Similarly, ADDM was 61.9, 68.1, 68.4 and 69.1% for groups A, B, C and D, respectively on Diet-2 with an improvement of 22.9, 4.6, 4.7 and 8.4% in ADDM when using supplementation with a fodder diet (Table VI).

TABLE V. APPARENT DRY MATTER DIGESTIBILITY OF PHASE ONE FODDER FEEDING AND PHASE FODDER FEEDING WITH A CONCENTRATE

Feeding Regime		Phase 1				Phase 2			
Group	Day	15	25	35	Average	60	70	80	Average
A	Intake (kg)	35.5	37.5	36.5	36.5	33.3	34.7	32.6	33.5
	Excreta (kg)	15.2	16.8	15.9	15.9	13.2	13.0	12.0	12.7
	Digestibility (%)	57.1	55.2	56.4	56.2	60.2	62.5	63.1	61.9
B	Intake (g)	65.2	67.6	65.5	66.1	61.4	59.9	64.7	62
	Excreta (Kg)	23.5	22.5	23.0	22.3	20.2	20.0	21.0	22.0
	Digestibility (%)	63.9	66.7	64.8	65.1	67.1	66.6	70.8	68.1
C	Intake (kg)	58.0	67.6	65.5	63.7	56.6	56.0	58	56.8
	Excreta (kg)	19.7	22.4	24.1	22.0	18.1	19.2	16.5	17.9
	Digestibility (%)	66.0	66.8	63.2	65.3	68.0	65.7	71.5	68.4
D	Intake (kg)	55.4	56	57.5	56.3	56	56.8	57.4	56.7
	Excreta (kg)	19.8	20.4	21.0	20.4	16.5	17.9	18.0	17.4
	Digestibility (%)	64.3	63.5	63.4	63.7	70.5	68.4	68.4	69.1

With the inclusion of concentrates in the diet digestibility improved by a maximum of 22.9% for group A whereas, group B's digestibility only improved by 4.6% (Table VI).

TABLE VI. AVERAGE DIGESTIBILITY'S FOR PHASE 1 AND 2 AND THE PERCENT CHANGE IN DIGESTIBILITY WITH THE CHANGE IN DIET

Group	Dry matter digestibility (%)		
	Phase 1	Phase 2	Improvement (%)
Group A	56.2	61.9	22.9
Group B	65.1	68.1	4.6
Group C	65.3	68.4	4.7
Group D	63.7	69.1	8.4

### 3.7 VFA profile, digestibility and methane production from fistulae animals

Digestibility and rumen VFA concentrations were measured on the fistulated animals and the results are shown in Table VII. Dry matter digestibility was found higher with the concentrate-supplemented group (Diet 2) than fodder group (Diet 1). With introduction of a supplement into the diet, a reduction in acetate to propionate ratio (A/P) of x% was achieved in both cattle and buffaloes. Acetate to propionate ratio was reduced with the inclusion of concentrates in the diet and the reduction was similar for both cattle and buffaloes. Methane production was estimated to be 350, 300 g methane per animal per day for cattle and 312 and 278g per animal per day for buffaloes fed Diet1 and Diet2, respectively. This represents a 14% reduction in methane production from cattle and a 10% reduction in methane production in buffaloes when the diet included concentrates.

TABLE VII. METHANE PRODUCTION, VFA PROFILE AND DIGESTIBILITY OF FISTULATED BUFFALOES AND CATTLE

Animal	Cattle		Buffalo	
	Diet1	Diet2	Diet1	Diet2
Feeding Regime				
Digestibility (%)	54.2 ± 0.8	61.7 ± 1.0	58.4 ± 1.4	64.1 ± 1.2
	Volatile Fatty Acids			
Acetate (%)	68.1 ± 1.9	66.3 ± 2.6	62.8 ± 2.6	60.0 ± 1.6
Propionate (%)	15.1 ± 0.8	18.8 ± 1.6	19.4 ± 1.1	23.4 ± 1.2
Butyrate (%)	11.2 ± 1.0	10.4 ± 2.0	13.0 ± 0.8	12.2 ± 2.4
Valerate (%)	2.3 ± 0.6	1.9 ± 0.8	3.0 ± 0.6	2.8 ± 0.4
Acetate/Propionate (A/P) Ratio	4.5	3.5	3.2	2.6
Reduction in A/P (over control)		27 %		26 %
	Methane CH <sub>4</sub>			
CH <sub>4</sub> g/animal/d	350	300	312	278
Reduction over diet1	14 %		10.8	

## 4. DISCUSSION

In Pakistan, there is a trend towards mixed farming with sorghum, millet and maize as common fodders in summer and with Berseem (*Trifolium alexandrinum*), oats and barley in winter [3, 20]. Animals are maintained in these mixed farming systems to produce milk and meat and for draft power purposes in cropping. The four animal groups comprising of Dajal young, Dajal adults, Sahiwal and Buffalo are representative of the ruminant livestock farming systems in Pakistan. The results found an additional gain of 75 kg in Buffalo calves with the supplemented feeding strategy when compared with the fodder alone strategy. There was an improvement of 49.7, 62.9, 38.5 and 32% in the feed to gain ratio of groups A, B, C and D, respectively with the supplemented feeding strategy compared to that of the fodder control. From a comparison of DM digestibility,



an improvement of 22.9, 4.6, 4.7 and 8.4% was evident when using a supplemented fodder ration than that of fodder alone. A possible explanation for better animal performance in supplemented feeding strategy could be that the rumen ecosystem requires balanced nutrients for optimum feed digestibility. When the available feed, like fodders, lacks some of the nutrients, digestion will be less efficient, lowering productivity and rearing methane production. Better performance in Buffalo than cattle could be attributed partly that buffalo could better utilize fibre fractions of feed than cattle [1, 21].

Forage with the addition of concentrate in the ration has an impact on the rumen fermentation and this improvement in formation will lead to a decline in the acetate to propionate ratio. Integrated approach of supplemented feeding resulted in 27% and 26% reduction in A/P ratio, respectively in cattle and Buffalo. Earlier workers [9] have also reported a reduction in A/P ratio with concentrate feeding. Methane production from simulation model estimated production of 350 and 300 g methane per animal per day in cattle and 312 and 278g per animal per day in buffalo on Diet1 and Diet2, respectively.

The reductions in methane production in this study are similar to those by Johnson and Johnson [8] who reported a similar reduction in methane production when concentrates were used in a forage based diets. Similarly, a decrease in methane production was reported when grass silage were supplemented with barley [14]. The supplement in present study also contained a high concentration of unsaturated fats, which can also result in decreased methane production. The results reported here are consistent with those reported by other workers [4, 7] who have reported a decrease in methane production with the inclusion of unsaturated fatty acids and these changes have often been associated with reductions in rumen protozoal numbers. Similar to our findings, earlier workers [6, 12] reported a decrease in rumen methane production with the fat inclusion in diets although with a mixed diet as used in this study it is not possible to differentiate the effects of fat from the improved nutritional status of the animals from the use of concentrates in the diets. It could be concluded that shifting fermentation pattern towards propionate from acetate with improved nutrition will meet the twin objectives of increased productivity and reducing the environmental impact of livestock.

## 5. CONCLUSIONS

The productivity of various classes of livestock could be minimized with integrated approach to ensure adequate feeding and this improvement in production will result in a reduction in methane production that will reduce the environmental impact of maintaining ruminant livestock.

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# **FARMYARD MANURE MANAGEMENT AND ITS EFFECT ON MAIZE FODDER AND SOIL NUTRIENTS**

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## **Abstract**

Stored manure in open heaps exposed to the sun, wind and rain accounts for substantial nutrient losses. A study was conducted to assess the effect of storing manures by traditional uncovered means or covered (under shade) and subsequently used as a fertilizer to determine effects on soil fertility and maize fodder performance. Manure from 5 animal groups was stored either traditionally or covered and the compost applied to a heavy loam soil. The DM and ash contents of the manure were higher in the uncovered compared to the covered manure. The N content of covered manure was 9.6 % higher along with P availability and exchangeable K than uncovered manures. Application of manure, whether covered or not, improved the fertilizer profile of soil than the non-manured control. There was 16.6% and 25.4 % increase in N in plots receiving manure from uncovered and covered manure piles respectively compared to the control plots that were not fertilized. The P level was increased by 6.8 and 19.3% in uncovered and covered manure plots compared to the control plots. Exchangeable K was increased by 1.3 and 1.6 times in uncovered and covered manured plots, respectively. Fresh fodder yield was 28, 32 and 35.2 metric tons per hectare for the control, uncovered and covered manure plots, respectively. Crude protein content was 8.58, 8.26 and 7.43 % respectively in fodders from covered, uncovered and non-manure plots. Overall, there was a conservation of nutrients in covered than in uncovered manure with a concomitant increase in fodder biomass yield.

## **1. INTRODUCTION**

Livestock is the second most important sector in Pakistan agriculture producing products that includes meat, milk, wool, bones, hair, fat, skin and hides. Pakistan's growing wealth in its major livestock industries is evident from the growth in livestock populations [19]. However, this growth needs to be sustained. Pakistan's soils are deficient in organic matter containing less than the threshold of 1.29% carbon needed for soil sufficiency. In a survey [4], carbon content in Pakistan soils varied from 0.52 to 1.38% across soil types and most soils contained less than 1% carbon. The low organic matter content of Pakistan soils is has been associated with climatic conditions, soil conditions, use of/lack of use of mineral fertilizers, poor economic conditions of farmers, intensive tillage, etc. Organic matter sources available for improving soil organic matter in Pakistan soils include livestock manure, crop residues, green manure, filter cake and silage, slaughter house waste, other solid and liquid based materials, compost and biogas compost, etc [10, 15].

Traditionally, Pakistani farmers have been concerned mainly with manures, also called farmyard manure, and these have been subjected to minimum manipulation by the farmers to improve the quality of the compost. Farmers keep livestock, which are tethered and fed using the cut-and-carry systems using assorted by-products of the annual

crops as fodders after harvesting the crop. Stalks of maize, sorghum or millet, straw from wheat or rice, and stover of groundnuts, chickpea and gram (mung beans) are commonly included in cattle feeds [24]. Crop residues are, therefore, not readily available for direct composting but are recycled instead, in the form of cattle manures. However, about half of all cattle/buffalo dung is used for household fuel and this trend to increase with the scarcity of other fuel sources. Manures traditionally receive a low priority from most farmers and storage this valuable asset is rather haphazard without much consideration of its potential use as fertilizer [21]. Storage of manure in rural households is usually in heaps exposed to sun, wind and rain, which can lead to substantial nutrient losses.

Manure is an important source of nutrients for crop production for the smallholder sector of Pakistan. It enables farmers to reduce inputs of commercial fertilizer, thereby, improving the profit margin of crop and pasture production. Nutrients contained in organic manures are released more slowly and are stored for a longer period in the soil, thereby, ensuring a long residual effect in contrast to chemical-based fertilizers [6, 23]. Although inorganic chemical fertilizers are used to replenish soil nutrients and increase crop yields, in most cases are too expensive for the smallholder sector. Integrated nutrient management strategies where both organic manure and inorganic fertilizers are used, have been suggested [20]. Maize is an important cereal crop within the tropics and is widely grown in Pakistan for fodder as well as for grain production. High palatability and adaptability along with adjustment in intercropping systems makes maize a first order crop choice for the tropics. Maize yield and composition are reported to be significantly altered with the application of manure [24].

The present study was conducted to compare manure managed using the traditional system compared with an improved manure management strategy and the effects tested on soil N/P/K profiles, maize fodder yield and fodder quality.

## 2. MATERIALS AND METHODS

Farmyard manure from experimental animals were collected, stored for four months and then applied to a heavy loam soil. Maize fodder was grown in the soil and effect of farmyard manure on fodder yield and nutritional value were determined as follows.

### 2.1 Experimental animals, manure collection and analysis

Five groups consisting of Sahiwal, Crossbred, Simmental, NiliRavi Buffalo and Desi cattle contained four castrate male animals per group were used to produce farmyard manure. The animals were 1-1.5 years old and fed on a 1:1 mixture of *Sorghum sudanese* and maize fodder. The groups were split into two subgroups of all the breeds and species with the manure from each subgroup being maintained in separate piles. Manure from the groups was stored as Conventional and Non-Conventional manure management systems (uncovered and covered, respectively).

Farmyard manure from animals in each subgroup was managed in according to two protocols. The Conventional manure management is the traditional practices of maintaining the piles as uncovered manure piles in open sun and without a cover. The Non-conventionally managed manure were kept in separate pits according the animal group with a concrete floor to receive urine from respective animal group via a drain and plastic sheets were spread over the pits to minimize N loss from these pits. The pits were sealed to avoid the leaching of stored manure and urine in both the treatments. Alternatively, manure managed conventionally was without plastic sheets. Manure collection from the groups continued for a period of four months. Samples from both the

treatments were taken by mixing thoroughly the manures from respective animal groups, taking representative samples for the two treatments and analyzed for dry matter (DM) content, ash, total N, available P and exchangeable K by standard methods as described in next section.

## **2.2 Application of farmyard manure and soil analysis**

The forage production experiment was carried out on a heavy loam soil irrigated with mix source of rain, ground and canal water, depending on availability. The forage experiment consisted of 24 plots of 5 x 5m (Marla) maize. There were two manure treatments and one control (NIL) contained 10, 10 and 4 plots respectively for Covered, Uncovered and NIL farmyard manure treatments. On the 20 plots of farmyard manure, manure was applied at the rate of 80 kg /plot while the NIL plots did not receive any manure. Surface soil samples were collected from all the experimental plots before and after the application of farmyard manure and analyzed as bulked samples. Farmyard manure was applied three weeks prior to planting and uniformly spread over the plots, followed by a light cultivation of the soil using a Chisel plough. All the agronomic factors were same for all the plots. No inorganic fertilizers were applied to any of the plots.

Nitrogen content of soil and farmyard manure samples were determined by Kjeldahl method as described by AOAC [2] using a semiautomatic Kjeldahl apparatus (VELP). Available P content of soil and farmyard manure samples was determined using the method of Olsen [17] as modified by Olsen and Somers [18]. Extractable K and exchangeable K content of farmyard manure and soil samples were determined by flame photometer (JENWAY) at 767 nm wavelength using the method of Ryan *et al.*, (2002) were:

$$\text{Exchangeable K (ppm)} = \text{Extractable K (ppm)} - \text{Soluble K (ppm)}$$

## **2.3 Cultivation of Maize fodder**

Maize fodder (variety Pak. Afghoy) was planted by broadcasting at the rate of 80 kg seed per hectare. Four irrigations were applied with the first irrigation being carried out at 21 day after sowing and subsequent applied after an interval of ten to twelve days according to water requirements for the maize crop. Plant height and fodder yield per plot were measured at 60 days of sowing. Chemical analysis of the fodder including DM, crude protein, ash and crude fibre content was performed using the methods of AOAC [1].

# **3. RESULTS**

## **3.1 Nutrient Profile of FYM**

The composition of farmyard manure stored using conventional and non-conventional methods is shown in Table I. Manure from 5 animal groups was mixed thoroughly and hence only two treatments were compared. The DM and ash contents were higher in uncovered farmyard manure than for covered farmyard manures. Nitrogen content of covered farmyard manure was higher than uncovered with an increase of 9.6 % in N content. Similarly, P availability and exchangeable K content was higher in covered than in the uncovered farmyard manure. The actively managed farmyard manure piles covered with a plastic sheet resulted in a 25% improvement of K content.

TABLE I. COMPOSITION OF FARMYARD MANURE STORED WITH AND WITHOUT COVERING

	DM %	Ash %	N %	P %	K %
Conventional (Uncovered)	52.04 ± 2.6	56.13 ± 1.1	0.94 ± 0.1	0.21 ± 0.03	0.20 ± 0.02
Unconventional (Covered)	49.81 ± 1.2	48.56 ± 1.5	1.03 ± 0.1	0.022 ± 0.05	0.25 ± 0.07

Each value is a mean value of duplicate samples ± shows standard error among mean values. FYM= N= Total Nitrogen; K= Available Potassium; P= Available Phosphorus.

### 3.2 Soil NPK profile as affected by FYM

The soil nutrient profile is presented in Table II. The soils receiving manure managed by two treatments or nil manure were compared. The experimental plots were on a heavy loam soil with 0.114%, 7.56 ppm and 34.8 ppm total N, available P and exchangeable K, respectively. On average, after the application of farmyard manure from the covered and uncovered manure treatments there was an improvement in the N, P and K soil profile. For N there was an increase of 16.6% and 25.4 % in plots receiving farmyard manure from uncovered and covered manure piles, respectively compared to plots that received no manure. The soil P level increased by 6.8 and 19.3% in uncovered and covered farmyard manure plots compared to the NIL plots. The highest changes occurred in soil mineral content with exchangeable K where increases of 1.3 and 1.6 fold was observed by the addition of uncovered and covered farmyard manure, respectively compared to the plots that were not fertilized with manure. The two manure treatments of covered and uncovered farmyard manure on average resulted in an improvement of 7.5%, 11.6% and 36.1% in N, P and K content in the soil compared with the non manure treated plots.

TABLE II: SOIL NUTRIENT CONTENT BEFORE AND AFTER THE APPLICATION OF FARMYARD MANURE FROM COVERED OR UNCOVERED STORAGE SYSTEMS

Nutrient Profile Treatment	N (%)		P (ppm)		K (ppm)	
	Uncovered	Covered	Uncovered	Covered	Uncovered	Covered
Control (Before FYM application)	0.114 ± 0.01	0.114 ± 0.02	7.56 ± 0.05	7.56 ± 0.04	34.8 ± 0.5	34.8 ± 0.1
After FYM application	0.133 ± 0.01	0.143 ± 0.01	8.08 ± 0.08	9.02 ± 0.04	80.46 ± 0.7	90.68 ± 0.3
Improvement over control	16.6%	25.4%	6.8%	19.3%	131.2%	160.5%
Improvement over uncovered	7.5%		11.6%		36.1%	

Each value is a mean value duplicate samples from two treatments mixed thoroughly ± shows standard error among mean values. FYM = Farm Yard Manure. N = Total Nitrogen; K = Available Potassium; P = Available Phosphorus.

### 3.3 Effect of FYM on fodder yield

Fresh fodder yield for the plots are provided in Table III. Average fodder fresh yield was 28, 32 and 35.2 metric tons/ha while DM yield was 3.9, 4.49 and 5.44 tons/ha for NIL, uncovered and covered farmyard manure plots, respectively. These yield differences are reflected in the sale of fodder at market rates, there the income was Pak rupees 42,000, 48,000 and 52,800 per hectare for NIL, uncovered and covered farmyard

manure plots, respectively. There was an improvement of 14.2% and 25% in uncovered and covered farmyard manure treated plots compared to the NIL plots.

TABLE III. EFFECT OF COVERED, UNCOVERED AND NIL FARMYARD MANURE ON MAIZE FODDER YIELD, ECONOMICS AND CHEMICAL COMPOSITION

Chemical Composition		Experimental Plots		
FYM Treatments		Nil FYM	Uncovered FYM	Covered FYM
Plant Height (m)		1.58 ± 0.09	1.67 ± 0.1	1.79 ± 0.02
Fodder Yield	Kg/plot	70 ± 1	80 ± 2	88 ± 2
	Tons/hectare* (calculated)	28	32.0	35.2
	DM Yield/hectare* (calculated)	3.9	4.49	5.44
Economics	Income/ha in Rs	42000/-	48000/-	52800/-
Proximate Analysis	DM (%)	13.94 ± 0.1	14.06 ± 0.13	15.48 ± 0.11
	CP (%)	7.43 ± 0.11	8.26 ± 0.11	8.58 ± 0.14
	Ash (%)	9.88 ± 0.13	10.11 ± 0.11	10.49 ± 0.13
	CF (%)	39.46 ± 0.1	39.20 ± 0.2	39.40 ± 0.1

Each value is a mean value of duplicate samples from plots receiving NIL, covered and uncovered manure. ± shows standard error among mean values. FYM = Farm Yard Manure; \* One Hectare= 10000 m<sup>2</sup>; Price of fodder@ Rs.1500/ metric ton

### 3.4 Proximate analysis of Maize fodder

The nutritional value of maize fodder grown on the plots is provided in Table III. The differences within each plot were not the scope of this study and only the treatment were compared. The dry matter content was the highest in covered manure plots and minimum in fodder from NIL plots. Crude protein content was 8.58, 8.26 and 7.43 % in fodders from covered, uncovered and NIL manure plots, respectively. A similar trend was observed in ash content of the fodders from the various manure treatments. When compared, the treatments of covered and uncovered, there was an improvement in nutritional value of the maize from covered farmyard manure plots compared to uncovered manure fertilized plots.

## 4. DISCUSSION

The nutrient profile of farmyard manures is affected by factors such as type of animal, feed and feeding supplements, bedding material used, the collection and storage system, as well as, the duration of storage and climatic conditions during storage. This study was conducted to measure the effect of storing manure and the subsequently effects of these storage methods on manure fertilizer quality when applied to soil for maize fodder production. Covering of the stored manure resulted in an improvement in chemical composition of manure compared to conventionally stored manures. This could partially be attributed to minimum losses of nutrients by sun, wind and rain in Covered FYM. The results confirmed our hypothesis that heavy rains, wind and open sun cause nutrient losses in conventionally stored manures. These results are consistent with the



results of other workers [12, 21] who have also reported a decline in nutrient profile of manures stored in the open and exposed to the environment.

The improved nutrient profile of the covered manures was also reflected in the improved fertilizer profiles of the soils treated with the covered fertilizer. Our results are in accordance with previous workers [7, 14] who reported an increase in soil nutrients for plots receiving manure as compared with control plots.

Fodder yield improved with manuring and improved manure quality. This improved yield increased the farmers' income per hectare through the increased quantity of hay available for sale. This assumes that there is no change in price with the improved N and other quality indicators for hay. These yield results are in accordance with those by Oad [16], who reported higher maize fodder yield by application of farmyard manure compared with the yield achieved for the control plots. However, fodder yield in our experiment was lower than those achieved with inorganic fertilizer in previous studies [3, 13]. This may be due to our experimental design where there was no use was made of inorganic fertilizers to assist the performance of the organic manure fertilizers. The sole use of organic fertilizers may have decreased fodder yield due to a long-term slow release of nutrients by manures as compared to the short term but quicker supply of nutrients by inorganic fertilizer. As well as yield improvements, there were also increases in dry matter content, ash and crude protein with the use of manure fertilizers. An improvement in nutritional value of fodder may be attributed to availability of required nutrients from soil in covered and uncovered manure plots compared to those available from that of nil manure-fertilized plots. A deficiency of major soil nutrients can lead to an altered ratio of leaves to stem which was recorded in this study. These findings are similar to those by Bilal [5] who found an improvement in fodder height and quality with the application of manure.

## 5. CONCLUSIONS

Management of manure from livestock in systemic manner resulted in an improvement in the fertilizer profile of manures. On use of such managed manure, there is an improvement in soil fertility and crop production as well as in nutritional quality of fodder grown on those soils.

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# INTEGRATED APPROACH FOR IMPROVING LIVESTOCK PRODUCTION USING INDIGENOUS RESOURCES AND CONSERVING THE ENVIRONMENT

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## Abstract

Two studies were conducted (i) to evaluate the voluntary feed intake, digestibility and performance of local Pyarzein growing calves fed on rice straw based diet supplemented with *Leucaena leucocephala*, *Gliricidia* and Rumex K-1 livestock fodder and (ii) to improve cattle production through reproduction using artificial insemination (AI) which is said to be the most reliable and applicable technique. In the first study, 4 local Pyar Zein growing calves averaging  $89.4 \pm 16.3$  kg live weight were allocated to 4 dietary treatments in a 4 x 4 Latin square design. Rumex K-1 is one of the promising fodders to supplement rice straw which could improve nutrient intake, digestibility and animals' performance. *Leucaena* and *Gliricidia* may also be useful N supplement in areas where cattle are fed on rice straw or low quality roughages. Fodder supplementation rate in this study (20% of total diet DM) did not improve DMI but it did improve digestibility. Fodder supplementation rate for rice straw based diet should be greater than 20% on a DM basis. However, issues of toxicity from secondary compounds contained in fodder leaves should also be accounted for. Crude protein content of the diet should be higher than 8% and roughage to concentrate ratio should be maintained at 50:50. In the second study, in 66 female cattle from 80 smallholder farms, the highest conception was in VS-3 (Pronounced Vulva Swelling), VR-3 (Redness in vulva), M-2 (Thick and hang down mucus) and UT-2 (Firm, tonic and muscular uterus) with the rate of 83, 92, 85 and 80% respectively. Highest conception rate (92.3%) was in cows inseminated at 40.6 to 41°C vaginal temperature and temperature above 39°C to 40.5°C also showed good conception rate (>80%). Improving animal productivity using better selection criteria for offspring from cross- breeding programme, calving rate was 79.1% and sex ratio was 43.7% (male) and 56.3% (female) respectively, total calf motility rate was 14.9%, in male 15.8% and in female 14.3% respectively.

## 1. INTRODUCTION

Livestock farming is important for the provision of food for the region and source of income for the poor farmers. In the developing countries, livestock production is below the potential due to many limitations such as inadequate feeding, mismanagement in reproduction, prevalence of diseases and lack of effective veterinary support. Reproduction is one of the most important considerations determining the profitability of cattle production both in dairy and beef animals. Artificial Breeding through Artificial Insemination and Embryo Transfer and its related techniques as a means of livestock improvement are now accepted and utilized worldwide [32]. Between those two technical approaches, Artificial Insemination is the most reliable and applicable technique to gain improved genetic potential of the animals. In Myanmar, the Livestock Development Project with the assistance of World Bank was implemented in 1976. In that project, livestock development programme and Artificial Insemination programme for cattle were undertaken simultaneously on a wider scale. The Livestock Breeding and Veterinary Department made many efforts for improving cattle production and productivity through the application of AI technique but little success was only achieved due to many

limitations. There is about 450 staff operating AI in 97 townships. Annual insemination ranged about 70,000 to 90,000 during the ten years (1976 to 1986). Average conception rate was recorded at 48.0% to 54.4%. AI performance has reached the peak in 1987 annual output usage from 90,000 to 140,000 and gradually declining during the last few years. 1995 to 2005 performances were slightly over 20,000. Insufficiency of liquid N, maintenance of upgraded and progeny tested bull, difficult in continuously import semen from abroad and with frequent exchange of animals among the small farmers are the constraints of the improvement.

Productivity and efficiency of livestock production per animal unit in developing countries is considerably less than in the more-developed world. Because forages and grasses for the ruminant livestock in the tropics are generally low in N and digestible nutrients [10]. Grasses in wet season and cereal straws in dry season are the major roughage sources for ruminants. Among the roughages, rice straw is the most abundant crop residue and used as a major source of roughage for cattle in Myanmar. Seasonal availability and high fluctuation price of protein rich livestock feeds such as oil cakes and cereal by-products also are limitations for smallholder farmers to meet the nutrient requirement of their livestock especially for ruminant animals. The utilization of cereal straws may be improved by supplementation with fodder tree foliages and legumes to improve the supply of fermentation N, carbohydrates and micronutrients ([4]). Dutta [8] also mentioned that foliage from legume trees offer an economic alternative to costly protein supplements. Forage from legume trees such as *L. leucocephala* and *Gliricidia sepium* can be used as N sources in supplementary feeds [12] and be used in combination with other locally available feed resources in formulating on farm supplementation [16]. *Gliricidia* has been grown very recently and Rumex K-1 livestock fodder is newly introduced livestock fodder in Myanmar. Therefore, it is necessary to study the response of leaf protein supplementation on live weight gain of growing calves.

In the previous work, using progesterone Radioimmunoassay technique and AIDA ASIA programme can evaluate the reproductive performance of the cattle. Project intervention consisted of two components; nutrition and reproductive management. Nutrition component of the intervention was based on the achievement of previous project RAS/5/035 in use of UMMB as feed supplementation has been carrying out as the first option and another second option is by using of locally available leguminous feed in cattle production. Project intervention on reproductive management was also based on the lessons learnt from the previous projects those were provided by IAEA such as, MYA/5/013 Improving smallholder dairy cattle production through improved reproduction and nutrition. The number of insemination per cow and conception rate at the selected project site was 1.28 and 78.08%, respectively. After the end of the project, although milk production was not significantly increased in the project sites, income from milk sold in individual herd was increased because farmers adopted and practiced the newly demonstrated feeding method and improved herd health programme.

The specific objectives of the project were to improve animal productivity and decrease emission of selected GHG (methane and carbon dioxide) and decrease leaching of selected nutrients (phosphorous and N) into the environment through feed manipulation and manure management strategies. Beside that project also aimed at improving livestock production using better selection criteria for offspring from crossbreeding programme, optimum utilization of appropriate indigenous cows, benchmarking for growth and reproduction and improving procedures for management, nutrition and health care programme for dairy farmers.

## 2. MATERIAL AND METHODS

### 2.1 A comparative study on the effects of different leaf protein supplementation on the performance of pyarzein growing calves fed rice straw based diet

The study was conducted at Livestock Research and Development farm, Livestock Breeding and Veterinary Department, Mingaladon Township, Yangon from 8 February to 21 May 2006. Four local Pyarzein growing calves, age between 12 to 18 months old were used to evaluate 4 dietary treatments during successive period. Their mean initial live weight before entering to the study was  $89.4 \pm 16.3$  kg. Before commencement of the trial, all animals were treated against internal and external parasites by Ivermectin 1% and Clorsulon injection at the rate of 50 kg per 1 ml (Ivocip plus, Cipla Ltd, India) and also were vaccinated against Haemorrhagic Septicaemia, Anthrax and O Type Foot and Mouth Disease (Recommended Vaccination Programme, Biologics Production Section, LBVD). Animals were confined in individual, well-ventilated stalls. One percent of live weight of fresh fodder leaf was offered to the animals in two equal portions before giving the rice straw (Table 1). Respective amount of concentrates (1% of live weight) were given at two equal parts before rice straw were fed all calves to ensure complete intake of concentrates. Rice straw was offered *ad libitum* by giving a weighed amount twice a day, providing 10% in excess of the previous day intake, at 8:00 hr and 14:00 hr so that there was always some left-over in the next feeding time. Amounts of all feeds offered and refused were monitored for calculating intake levels. Clean water was provided free choice to all animals. Animals were weighed before morning feeding at the end of every trial on three successive days. Digital weighing machine for cattle was used to determine the live weight of animals. The amount of supplementation in each trial was adjusted based on live weight. Each feeding trial consisted of 5 days preliminary, 15 days experimental and last three days during experimental period for sample collection.

The growing calves were randomly allocated to receive 4 dietary treatments according to 4 x 4 Latin Square design. Table 1 shows the experimental design of dietary treatments.

TABLE I. EXPERIMENTAL DESIGN

Calf No.	Period 1	Period 2	Period 3	Period 4
1	RSLC	RSRC	RSGC	RSC
2	RSC	RSLC	RSRC	RSGC
3	RSRC	RSGC	RSC	RSLC
4	RSGC	RSC	RSLC	RSRC

RSLC: Rice Straw + Leucaena + Concentrate; RSRC: Rice Straw + RumexK-1 + Concentrate; RSGC: Rice Straw + Gliricidia + Concentrate; RSC: Rice Straw + Concentrate. Calf concentrates consisted of rice bran, pea bran, sesame cake and chickpea husk with the proportion of 30, 20, 30 and 20% respectively as fresh basis. Fodder leaf was harvested every day and directly fed to animals as fresh basis.

Dry matter (DM) and Crude Protein (CP) in feed and faeces were determined according to the methods of [2]. Neutral Detergent Fibre (NDF) and Acid Detergent Fibre (ADF) were determined by the method of [9]. Data were subjected to analyze by the Analysis of Variance and treatment means were compared by using Duncan's new multiple range Test [18].

## 2.2. Study on the basic indices used for improving conception rate on artificial insemination of dairy cattle at field level

The study was conducted in Mingaladon and North Okkalapa Township during the period from February 2005 to February 2007 (24 months). Villages involved in the study area were situated along the No. 3 main high way road and those villages are approximately 9 to 18 miles far away from city centre of Yangon.

### 2.2.1 Farm characteristics

All the animals used in present study were crosses of Friesian blood with 25, 50 and 75 percent. 166 dairy females (31 heifers and 135 cows) from 80 smallholder farms with holding size ranging from 2 to 24 of adult female cattle were involved in the study. Heifers were age between 2 to 4 years and cows were 4 to 9 years with 1 to 6 parity. All dairy farmers practice cut and carry feeding system and two times milking per day. Dairy animals in all farms were tied at stanchion all the time. Animals were fed on roadside grass and rice straw at *ad libitum* basis in rainy to mid winter (June to December) and late winter to summer (February to May), respectively. Ingredients to make homemade concentrates were purchased locally and they were formulated to form dairy concentrates by different recipes according to their preferences. Some farmers prefer Artificial Insemination technique only to have their animals to be pregnant and some farmers use Artificial Insemination as alternative tool to conceive their animals where breeding bulls were not available. All farms involved in this study were practiced both Artificial Insemination and natural breeding.

### 2.2.2 Semen quality control

Frozen semen straws produced from 4 breeding bulls at AI station were used for Artificial Insemination in both of field works and research purposes. The identification number of those bulls were Fa 15 (50% local Friesian), Fc19 (75% local Friesian), F10 (100% local Friesian) and F20 (100% Local Friesian). Frozen semen straws used in this study were inspected to pass quality control procedure according to [27] and certified for AI. Post thawing ability of frozen semen straws (Q.C. Passed) are shown in Table II.

TABLE II. POST THAWING ASSESSMENT OF FROZEN SEMEN OF DIFFERENT BREEDING BULLS (2004 - 2005)

Parameter <sup>1</sup>	Fa15	Fc19	F10	F20	Acceptable range for AI/ 0.25 CC straw <sup>2</sup>
Assessment Freq:	12	68	26	34	
Motility % (Active Forward Movement)	37.1±2.4	39.6±3.7	37.0±1.6	36.7±1.5	30
Motile sperm x 10 <sup>6</sup>	12.5±1.4	13.1±1.5	12.9±0.6	12.4±1.1	7.5-10
Total Abnormal (%)	2.0-8.0	3.6-9.0	4.0-8.5	5.0-11.0	5% maximum of any defect and 20% total abnormal

Source: (1) Quality Control Assessment record, frozen semen production laboratory, Livestock Research and Development Section, LBVD, Yangon.

(2) Reproduction in farm animals, Department of Veterinary Clinical Science and Animal Husbandry, The University of Liverpool, U.K.

### 2.2.3 Heat detection

On every farm visit, farmers were asked to provide reproductive history of female cattle that were going to be received AI. Farmers have been trained to observed detection of oestrus minimum of four times per day. The time of oestrus when farmers have started noticed was also recorded. Visual examination and per rectal examination were done by researchers before giving AI. Uterus was examined for size and consistency. Table III shows the indices used to confirm female dairy cattle in oestrus.

TABLE III. INDICES USED TO CONFIRM FEMALE DAIRY CATTLE IN ESTRUS

Indices	Vulva swelling (VS)	Vulva redness (VR)	Mucus (M)	Uterine tone (UT)
Score 1	Slight	Pale pink	Very little, watery	Bigger than normal, flaccid
Score 2	Moderate	Pink	Thick and Hanging down	Firm, tonic and muscular
Score 3	Pronounced	Red	Copious, hanging down to ground	Very firm and more muscular

### 2.2.4 Artificial Insemination

All animals in this study were inseminated with frozen semen straws produced during the period from 2004 to 2006. All inseminations were performed under shade to avoid direct sunlight on to the AI gun (semen straw loaded). AI gun and frozen semen straw were prepared systematically and introduced gently through the cervix under control of the hand in the rectum. Some semen dose was deposited just in front of the internal opening of the cervix into the body of uterus (cervical uterine junction) [27] and a small amount of semen was drawn back into the cervix and deposited [24]. After withdrawal of the gun the vulva was massaged to assist sperm transport. Intra-vaginal (close to external opening of cervix) temperature was taken by digital thermometer after insemination.

### 2.2.5 Recording

Name of animal's owner, address, cow's ID, breed, percentage of Friesian blood, age, parity, BCS, days in milk, daily milk yield around insemination, date of first oestrus after calving, previous reproductive history, time of oestrus started, time of insemination, vulva swelling, redness, mucus, uterine tone, vaginal temperature and straw identifications were recorded in every farm visit. All data were stored at computer database programme.

### 2.2.6 Pregnancy diagnosis

Heifers and cows that did not come back into oestrus after three months of AI were tested for pregnancy by rectal palpation as described by [32].

### 2.2.7 Statistical Analysis

Study data were analyzed for statistical significance by using Goodness of Fit Test described by [31].



### 2.3. Improving animal productivity through the use of better selection criteria for offspring from cross- breeding programme

Mostly exotic crossbreed and some dual purpose Pyazein cows can used for milk production under partly grazing and partly feeding management conditions. For the selection of the superior young bulls, the study was done in Indigenous bull mother farm of Research and Development section with (6) numbers of indigenous pyazein cows. 104 cows from another three Pilot Farms situated near Research and Development Section, Mingaladon Township also keep a close watch on achievement of bull calves candidate. 104 cows from the pilot farms were crosses of Friesian with 25, 50, 75%.

Pyazein indigenous cows produced 5 to 6.7 kg/day and Friesian crosses cows were produce 10 kg to 24 kg milk and has a herd average of 10.8 kg/day. All breeding cows had their records of sire and dam performances. A.I only was used in Research farm. Both Artificial Insemination and natural mating were used in other three pilot farms. Frozen semen straws produced from 4 breeding bulls from AI station and Foreign Friesian straws imported from Semex Canada, France and China were used for Artificial Insemination. Eight bulls with record of performance from the pilot farms were also used for natural breeding. This programmed was started in February 2005.

### 2.4 Method for estimating cost and benefit for young bull.

Total cost of rearing a young bull to 2 years of age	= Concentrate + Roughage = 221625 + 93000 = 3146255 kyats
The price for a young bull at 2 years of age	= 600000 kyats
Profit	= 600000 – 314625 kyats = 285375 kyats = 285 US\$ ( 2 yrs of age) = 143 US\$ / 1year
Price of quality breeding bull at 3 to 4 years of age	= 11,00000

## 3. RESULTS

### 3.1. A comparative study on the effects of different leaf protein supplementation on the performance of pyarzein growing calves fed rice straw based diet

#### 3.1.1 Chemical composition of feedstuff and experimental diets

The chemical composition of rice straw, Leucaena, Gliricidia, Rumex K-1 fodder and concentrates are shown in table IV.

TABLE IV. CHEMICAL COMPOSITION OF RICE STRAW, TREE FODDERS AND CONCENTRATE (% DM BASIS)

Name of sample	%DM basis				
	DM	OM	CP	NDF	ADF
Rice straw	97.50	76.20	3.56	66.5	49.2
Leucaena leaf	22.36	18.15	22.10	32.1	15.8
Gliricidia leaf	30.05	5.14	21.32	65.6	35.7
Rumex K-1 leaf	6.38	3.25	29.94	69.2	28.5
Concentrate	71.32	67.87	16.70	60.3	25.7

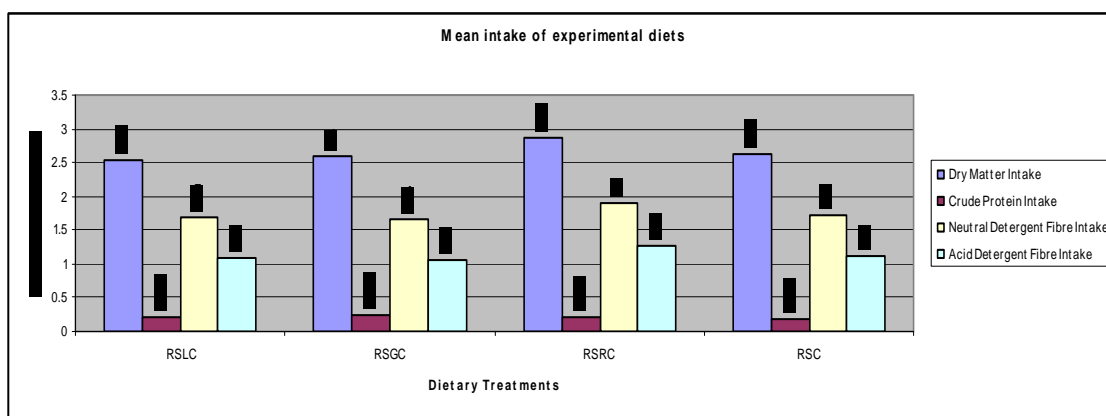


FIG. 1. Mean Intakes of DM, CP, NDF and ADF of the 4 dietary treatments

Each of experimental diet contained a kind of fodder as 1% of live weight of animal as fresh basis. Dry Matter content of all experimental diets except no fodder-supplemented diet (RSC) was almost similar. Crude Protein contents of RSLC, RSRC, RSGC and RSC were 8.6, 9.6, 7.3 and 7.1% respectively. NDF and ADF contents also were similar in all experimental diets. Chemical composition of experimental diets was shown in table V.

TABLE V. CHEMICAL COMPOSITION OF DIETS

Name of sample	% DM basis				
	DM	OM	CP	NDF	ADF
RSLC	71.0	69.2	8.6	62.0	40.3
RSGC	69.6	72.5	7.3	65.0	43.0
RSRC	70.6	65.1	9.6	64.6	40.7
RSC	88.7	73.9	7.1	64.8	42.8

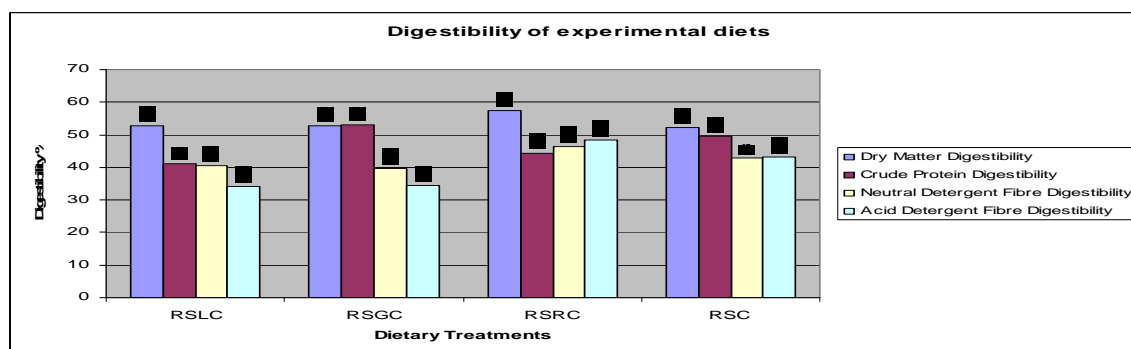


FIG. 2. Apparent digestibility of DM, CP, NDF and ADF of the 4 dietary treatments

### 3.1.2 Palatability order of the different type of fodders

According to the selection behaviour of the animals, it was observed that *Leucaena* leaf supplementation was consumed within short period (<15 minutes) and followed by *Gliricidia* and *Rumex K-1* (>30 minutes). Table VI shows the palatability order of different types of fodders.

TABLE VI. PALATABILITY ORDER OF THE DIFFERENT TYPE OF FODDERS

Sr	Name of fodder	Palatability Ranking	Time taken to consume
1	Leucaena	1	< 15 minutes
2	Gliricidia	2	20 to 30 minutes
3	Rumex K-1	3	>30 minutes

### 3.1.3 Voluntary dry matter intake and Crude protein intake

Table VII shows mean values of daily dry matter intake, crude protein intake and digestibility of different fodder supplemented diets. Dry matter intake of RSLC, RSGC, RSRC and RSC calculated based on 100 kg BW were 2.53, 2.60, 2.87 and 2.64 kg/day respectively. However daily dry matter intakes of different fodders were numerically differed, there were no significant differences observed. Mean values of daily crude protein intakes of RSLC, RSGC, RSRC and RSC were 226, 246, 213 and 188 g/day respectively.

### 3.1.4 Dry matter digestibility and crude protein digestibility

Dry matter digestibility for RSLC, RSGC, RSRC and RSC are shown in Table VII. Highest DM digestibility was observed significantly in Rumex K supplemented diet. Crude protein digestibility for RSLC, RSGC, RSRC and RSC are shown in Table VII. Highest CP digestibility was observed in RSGC diet but all dietary treatments were not significantly differed. Table 5 also shows the digestibility of experimental diets.

### 3.1.5 Neutral Detergent Fibre and Acid Detergent Fibre intake

Intakes of Neutral Detergent Fibre for RSLC, RSGC, RSRC and RSC are shown in Table VII. Numerically highest NDF intake was observed in Rumex K-1 containing diet (RSRC) but NDF intake in all diets were not differed significantly. Acid Detergent Fibre intakes for RSLC, RSGC, RSRC and RSC diets are shown in Table VII. ADF intakes were not significantly differed among experimental diets.

### 3.1.6 Neutral Detergent Fibre digestibility and Acid Detergent Fibre digestibility

The digestibility of NDF of RSLC, RSGC, RSRC and RSC diet are shown in Table VII. While in digestibility of Acid Detergent Fibre, significant differences ( $P < 0.05$ ) were observed in all experimental diets. RSRC and RSLC diet shows highest (48.4%) and lowest (34.2%) respectively. ADF digestibility of RSLC diet was similar to those of RSGC diet. RSRC and RSC diet also shows similar ADF digestibility.

TABLE VII. VOLUNTARY DRY MATTER, CRUDE PROTEIN INTAKE AND DIGESTIBILITY (100 KG LIVE WEIGHT)

Parameter	Diet				P value
	RSLC	RSGC	RSRC	RSC	
DM Intake (kg)	2.53	2.60	2.87	2.64	NS
DM Digestibility	52.8 <sup>a</sup>	52.7 <sup>a</sup>	57.4 <sup>b</sup>	52.3 <sup>ac</sup>	P < 0.05
Crude Protein Intake (g)	226	246	213	188	NS
CP Digestibility	41.1	53.1	44.3	49.5	NS
NDF intake (kg)	1.69	1.66	1.90	1.71	NS
NDF Digestibility	40.49	39.8	46.26	43.00	NS
ADF intake (kg)	1.09	1.05	1.26	1.13	NS
ADF Digestibility	34.19 <sup>a</sup>	34.51 <sup>a</sup>	48.44 <sup>bc</sup>	43.17 <sup>b</sup>	P < 0.05

<sup>a, b, c</sup> means bearing different superscripts are significantly different ( $P < 0.05$ )

### 3.1.7 Roughage to concentrate ratio and inclusion rate of fodder leaf in total diet

Roughage to concentrate ratio of RSLC, RSGC, RSRC and RSC diets are shown in Table VIII. All diets except RSC diet used in this study were almost similar (approximately 50:50). Fodder leaf proportion to the total diet as DM basis in RSLC and RSGC diet were 21:79 and 27:73 respectively where as in RSRC diet was 9:91 but RSC diet was 0:100 due to absence of fodder supplementation in RSC diet. Table VIII shows Roughages to concentrate ratio and inclusion rate of fodder leaf in the total diet DM of experimental diets.

TABLE VIII. ROUGHAGES TO CONCENTRATE RATIO AND INCLUSION RATE OF FODDER LEAF IN THE TOTAL DIET DM OF EXPERIMENTAL DIETS

Parameter	Diet			
	RSLC	RSGC	RSRC	RSC
Roughage: Concentrate	49:51	49:51	45:55	37:63
Fodder leaf, % of total diet DM	21	27	9	0

### 3.1.8 Average Daily Gain (ADG) and Feed Gain Ratio (FGR)

Average Daily Gains on calves receiving different fodder supplemented diets were shown in Table IV. ADG of RSLC, RSGC, RSRC and RSC were not significantly different. Percent live weight gains of calves fed on different diets of above order were 0.46, 0.41, 0.50 and 0.41. Feed gain ratio for RSLC, RSGC, RSRC and RSC were 5.33, 6.35, 5.97 and 6.35 respectively.

TABLE IV. AVERAGE DAILY GAIN AND FEED GAIN RATIO

Parameter (Mean value)	Diet				P value
	RSLC	RSGC	RSRC	RSC	
Initial live weight (kg)	99.68	100.80	99.55	99.93	NS
Final live weight (kg)	109.06	109.05	109.13	108.25	NS
Live weight change (kg)	9.38	8.25	9.58	8.33	NS
Days of measurement	20	20	20	20	
ADG (g)	468.75	412.50	478.75	416.25	NS
ADG, % LW	0.46	0.41	0.50	0.41	NS
DMI (kg/day)	2.50	2.62	2.86	2.64	NS
FGR (kg DMI/kg ADG)	5.33	6.35	5.97	6.35	NS

### 3.2. Study on the basic indices used for improving conception rate on artificial insemination of dairy cattle at field level

The results obtained in 166 cows subjected to AI are summarized in Table X.

#### 3.2.1 Vulva swelling, redness and conception rate-

Highest conception rate were found in animals those had pronounce (highest) vulva swelling (VS-3) and more redness of internal lining of vulva lips (VR-3) during insemination. Eighty three percent and 91.94% conception were found in highest vulva swelling and more intensity of redness respectively while in slight swelling of vulva and pale pink colour vulva owned animals during insemination showed lowest conception rate, 53.33 and 63.16% respectively. Number of conception rate owing to different scores of vulva swelling and redness during insemination was highly significant ( $P < 0.005$ ).

#### 3.2.2 Mucus strand, uterine tone and conception rate

Mucus strand, which were thick and hanging down (M2) and good uterine tone (UT2) could provide highest conception rate of 84.75 and 80.36% respectively. Table 3 showed the oestrus parameters and its relation to conception rate. Number of conception owing to different scores of mucus strand and uterine tone during insemination was highly significant ( $P < 0.005$ ).

TABLE X. VULVA SWELLING, REDNESS, MUCUS AND UTERINE TONE RELATION TO AI CONCEPTION RATE

Parameter	Conception rate (%)			P value
	Score 1	Score 2	Score 3	
Vulva swelling	53.33	75.94	83.08	$P < 0.005$
Vulva redness	63.16	71.21	91.94	$P < 0.005$
Mucus	70.75	84.75	79.17	$P < 0.005$
Uterine tone	54.55	80.36	74.44	$P < 0.005$

#### 3.2.3 Time of insemination and conception rate

Highest conception rate (93%) was found in animals inseminated after 12 to 14 hours after onset of heat. Animals inseminated at 14 to 24 hours after onset of heat also were showed good conception rate (>75%).

TABLE XI. TIME OF INSEMINATION AND CONCEPTION RATE

Parameter	Time of insemination after onset of oestrus (hours)								P value
	08-10	10-12	12-14	14-16	16-18	18-20	20-22	22-24	
No. of Insemination	6	50	15	40	40	28	9	12	
No. of Conception	4	32	14	31	34	21	7	9	$P < 0.01$
Conception rate (%)	66.7	64.0	93.3	77.5	85.0	75.0	77.8	75.0	

#### 3.2.4 Vaginal temperature and conception rate

During 12 to 24 hours after onset of oestrus, the animals showed vaginal temperature range between 38.0 and 41.0°C. Some animals have reached vaginal temperature at 41.0° C before 15 hour after onset of oestrus and it was sustained until 18 hours after onset of oestrus. It was found that vaginal temperature of all animals was not

less than 38°C and not beyond than 41°C during oestrus period. Majority of animals in oestrus showed vaginal temperature at 38.5 to 39.5°C and they were coincided with uterine score 2 to 3. Highest (92.3%) and lowest conception rate (64%) were found in highest vaginal temperature (40.6 to 41.0°C) and lowest temperature (38°C) respectively.

TABLE XII. INTRA-VAGINAL TEMPERATURE RELATION TO AI CONCEPTION RATE

Parameter	Intra-vaginal temperature (°C)							P value <i>P</i> < 0.05
	38.0- 38.5	38.6- 39.0	39.1- 39.5	39.6- 40.0	40.1- 40.5	40.6- 41.0	41.1- 41.5	
No. of Insemination	25	37	43	49	19	13	6	
No. of Conception	16	25	36	37	17	12	6	
Conception rate (%)	64 <sup>a</sup>	67.6 <sup>b</sup>	83.7 <sup>c</sup>	75.5 <sup>d</sup>	89.5 <sup>e</sup>	92.3 <sup>f</sup>	100 <sup>g</sup>	

<sup>a, b, c</sup> means bearing different superscripts are significantly different (*P* < 0.05)

### 3.3. Improving animal productivity through the use of better selection criteria for offspring from cross- breeding programme

After breeding (110) Cows from (4) pilot farms with AI and natural mating, there are (87) Number of calves were borne. 38 were male calves and 49 were female calves. Calving rate was 79.09% and sex ratio was 43.68% (male) and 56.32% (female) respectively.

Six male and 7 female calves died within a year. Overall calf motility rate was 14.9% or 15.8% in male and 14.3% in female, respectively.

From the Research and Development farm, 7 cows gave birth to 16 calves (6 male, 10 female) over a 4-year period. Calf growth rates ranged from 0.15 to 0.82 kg/day - recorded 32 young male calves in 2007.

TABLE XIII. CALVING RATE (CR) ON 4 CATTLE FARMS IN 2006

Farm name	Number of cows	Number of male calves born	Number of female calves born	Total calves born (% CR)
TM farm	25	9	11	20 (80%)
Silver pearl	20	6	8	14 (70%)
Shwepuzune	59	20	27	47 (79.7%)
Rand D farm	6	3	3	6 (100%)
Total	110	38	49	87 (79.1%)

SELECTED YOUNG BULLS FROM THE 4 PILOT FARMS IN 2008

Bull ID	Date of birth	Breeding	Sire Breed	Dam breed	BCS
SPZ 001	6.4.06	AI	Friesian100% Aquann Valiant Char	50% F	3.5
SPZ 002	3.2.06	AI	China 100%F	50% F	3.5
SPZ 003	2.10.06	AI	China 100%F	50% F	3
TM 1	17.2.06	Bull	Garmani 50%F	50%F	3.5
TM 2	11.5.06	Bull	Garmani50% F	75%	3.5
RandD 01	5.11.06	AI	F20 100%F	Pyazein 485	3
RandD02	10.6.06	AI	F20 100%F	Pyazein 486	3
SP -5	10.12.05	Bull	SPZ 62.5%F	75%F	3

First time selection was based on the phenotype and good genetic merit at the age of one year. (16) Numbers of young bulls were selected as young sire bulls. At the age of

1½ to 2 year in 2008- total (8) number of young bulls were second time selected based on the physical appearance, body structure, and growth rate and some in semen quality for the purpose to be proven sire. For the next 3.5 to 4 years, the superior bulls are to be selected according to the milk recording of heifer daughters. By that time, we would hope two or three proved superior young bulls could be selected for the department and the rest of the other young bulls and their frozen semen could be used as draught cattle and discarded. The diagram of the selection of superior young bulls is as follow.

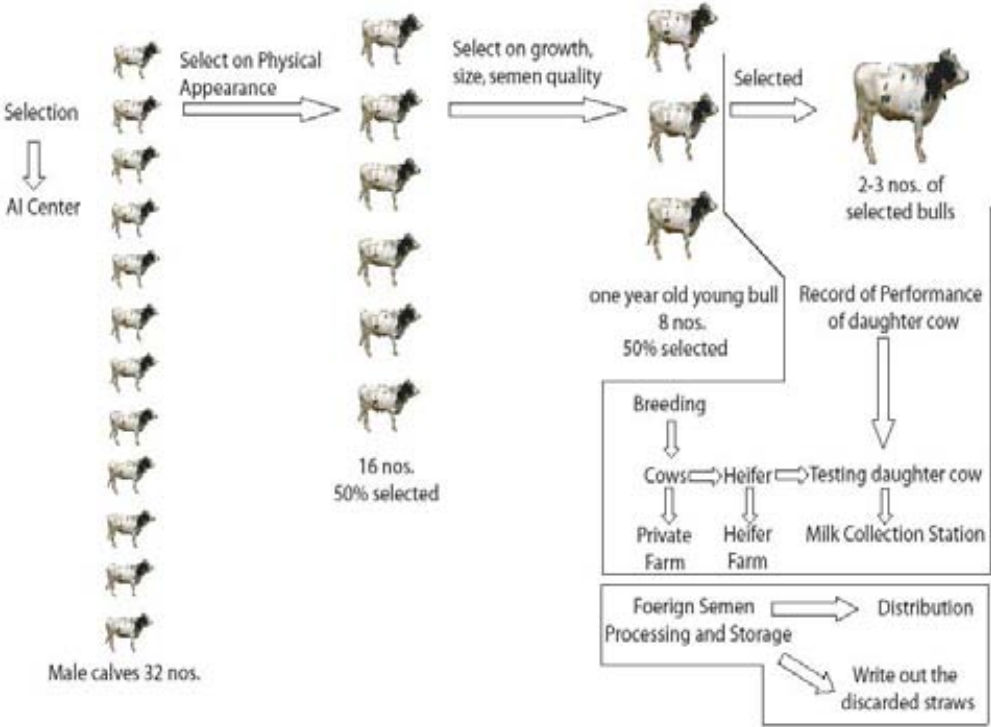


FIG 3. Ways of selecting the dairy cattle proven sire Bulls

Selected 8 young bulls from 4 pilot farms in the project area. The cost of feeding the calves from weaning to 2 years of ages is calculated below.

- CONCENTRATE FEEDING

Age (Month)	Feed consumption (Concentrate)	Feed price per kg (In Kyat )	Cost in Kyats
2 - 3	1/2 kg per day	225/kg	$1/2 \times 30 \times 1 \times 225 = 3,375$
3 - 5	2/3 kg per day	225/kg	$2/3 \times 30 \times 2 \times 225 = 9,000$
5 - 12	1 kg per day	225/kg	$1 \times 30 \times 7 \times 225 = 47,250$
12 - 24	2 kg per day	225/kg	$2 \times 30 \times 12 \times 225 = 162,000$
For 2 Years			Total 221,625

## - RICE STRAWS FEEDING

Age (Month)	Feed consumption (Rice Straw)	Feed price per kg ( In Kyat )	Cost in Kyats
4 - 6	5 kg per day	10/kg	$5 \times 30 \times 2 \times 10 = 3,000$
6 - 12	10 kg per day	10/kg	$10 \times 30 \times 6 \times 10 = 18,000$
12 - 24	20 kg per day	10/kg	$20 \times 30 \times 12 \times 10 = 72,000$
For 1 1/2years			Total 93,000

(30 = days/ month) (2= for 2 months) (6= for 6 months) (12= for 12 months)

## 4. DISCUSSION

### 4.1. A comparative study on the effects of different leaf protein supplementation on the performance of pyarzein growing calves fed rice straw based diet

#### 4.1.1 Chemical composition of feedstuff and diets

The dry matter content of rice straw used in this experiment was 97.5% and which was relatively higher than those of 91.2% [23], 93.9% [19] and 95.2% [22] but Organic Matter content (76.2%) was lower than the value (82.0%) reported by [19], (83.5%) reported by [23] and (83.7%) reported by [22]. The crude protein content of rice straw used in this study was 3.56% and that was relatively higher than those of 3.27% [23], reported value of 2.5% [19] and 2.6% [21]. The different chemical composition of rice straw indicated the differences among rice varieties, storage method and environmental factors. Rice straw used in this experiment was stacked in open air and fed to animals directly. The experiment was carried out during the hottest months of the year. For those reasons, the DM content of rice straw in this experiment was relatively higher than those of reference values. The straws Neutral Detergent Fibre (NDF) and Acid Detergent Fibre (ADF) contents were comparable to other published values [21, 22]. Mean Crude protein (CP) content of *Leucaena* leaf in this study was 22.1% and which was similar to values reported by [14] and [5] but lower than 29.6% value reported by [8]. Mean value of CP for *Gliricidia* was 21.32% and which was lower than observation of Ni Ni Maw [14] (18.64%) and 14.7% reported by [5]. Crude Protein content of Rumex K-1 used in this study (29.94%) was felt within the value of product specifications, 28 to 34% CP at DM basis (<http://www.romax.com.cn>). Higher and lower CP content were found in RSRC and RSC diet, which may be due to different CP content of fodder leaves included in the diets.

#### 4.1.2 Palatability of fodder

Palatability of tree fodder has been attempted to assess through short-term intake trials. These trials do not take into consideration the fact that animals can be trained to consume fodder, which they do not like the first day they are exposed to it. Palatability of tree fodders in this study was assessed by period needed to consume specific type of fodder by animals completely. Of the tree fodders, *Leucaena* had highest palatability but its containing diet showed numerically low DMI. Reluctance to eating *Gliricidia* leaves may be due to its odour ([3], 1986), astringency and post-negative influences of tannins on the epithelium of oral cavity of the animals [1]. Lowry [11] also mentioned that in some parts of the world the use of *Gliricidia* is limited for ruminants by unpalatability. However Rumex K does not contain tannin according to product specification



(<http://www.romax.com.cn>) and it was observed that cattle taken some times to complete consumption of Rumex K-1 (<30 minutes) and it may be due to its sour taste.

#### 4.1.3 Voluntary dry matter intake and crude protein intake

Total dry matter intake of Rumex K-1 supplemented diet in the present study was higher (2.87 kg/day) than in the other treatments calculated based on 100 kg live weight but not differed significantly. NRC [15] recommended that thin yearling steers should be provided high quality roughages at the rate of 3% of live weight as dry matter basis. Voluntary dry matter intakes on this study were slightly lower than (<3%) [15] recommendation but it means that animals fed only on high quality roughages. In this study animals were supplemented with green fodders as well as concentrates. It is therefore DMI may be lower than [15] due to substitution of concentrate to roughages. Another reason for lower DMI compared to [15] was due to animals suffered from heat stress during experimental period, which was summer in Myanmar. Leng [10] also found that in tropical countries, often low intakes of poor quality forage by ruminant may be imposed by a combination of metabolic heat stress, high environmental temperature and humidity. Preston [17] stated that 20% of the dry matter of a diet in the form of green forage is enough to meet the requirement for microbial nutrients. Green fodder supplementation rate for RSLC and RSGC diets in this study were agreed with them while in RSRC diet, it was very much lower than their recommendation. On the other hand, fodder supplementation rate in this study was contrary to [6] and who suggested that optimum dietary level on DM basis of leguminous fodder should be 30 to 50% for practical application.

To maintain the live weight of cattle, high level of forage tree supplementation at 1.1% of live weight or 33% of total diet DM of either *Leucaena* [13] or *Gliricidia* [7] on rice straw based diets are required. Palatability can be an important factor for the voluntary intake of fodder (Ralph, 1999) however, in this study, palatability could not determine the voluntary intake or DMI of all experimental diets may be due to lower supplementation rate of tree fodder (>0.3% Live weight) than recommendations made by other workers. Due to very low dry matter content (6.38%) of Rumex K-1 and based on the feeding level (1% of live weight of fresh basis), its dry matter intake was very much substituted with rice straw (9:91) to maintain required DMI for animals themselves. Devendra [5] stated that voluntary feed intake is influenced a very large extent by the dietary crude protein content. A low protein and high fibre content are major limitation for the utilization of rice straws. Crude protein content of different fodders used in this study was higher than 20% and could be used as protein supplement to improve rice straw utilization. Numerically highest DMI was observed in RSRC diet may be due to its highest CP content.

Numerically highest CPI was observed in RSGC diet but treatment means were not significant. Although, Rumex K-1 provided highest CP (29.94%) to the diet, CPI for it containing diet showed lower than RSLC and RSGC due to its lower supplementation rate at DM basis. Figure 1 shows the mean intakes of DM, CP, NDF and ADF.

#### 4.1.4 Dry matter digestibility and crude protein digestibility

Apparent digestibility (%) of DM and ADF exhibited a higher trend ( $P < 0.05$ ) in Rumex K-1 supplemented diet resulting in higher dry matter intake than in other treatments. Preston [17] mentioned that diet less than 8% CP are considered deficient, as they cannot provide the minimum ammonia levels required for microbial activities in the rumen. Rumex K-1 does not contain tannins, highest CP content of its containing diet

(9.6%) compared to other diets and it will be rapidly degraded in the rumen, providing high level of NH<sub>3</sub>-N, which favoured to increase microbial activities and promote digestibility of its supplemented diet. In the RSLC and RSGC diets, *Leucaena* and *Gliricidia* respectively contained tannin and which bound to plant protein. Tannin determines leaf protein to be digested slowly and provide lower ammonia N into the rumen insufficiently for optimum microbial activities, which could lower digestibility. However, *Leucaena* and *Gliricidia* may contain different levels of tannin, crude protein digestibility of all experimental diets were not differed significantly due to there was no significant differences in DMI, CPI and CP content of the diets. Figure 2 shows the apparent digestibility of 4 dietary treatments.

#### 4.1.5 Neutral Detergent Fibre and Acid Detergent Fibre intake

NDF intakes of all experimental diets in this study were not observed significantly. It may be due to NDF contents of different leaf protein sources except *Leucaena* were similar and amount fed to animals were the same (calculated on the 1% of live weight) because live weight of experimental animals were almost the same. Even though highest and lowest ADF content were observed in Rice straw and *Leucaena* leaf respectively, Acid Detergent Fibre intakes of all experimental diets were not differed significantly. Possible reason for similar intakes of NDF and ADF on experimental diets was lower supplementation rate (>0.3% of live weight at DM basis) of green fodder which contributed to lower intakes.

#### 4.1.6 Neutral Detergent Fibre and Acid Detergent Fibre digestibility

Digestibility of Neutral Detergent Fibre of RSLC, RSGC, RSRC and RSC diet were not differed significantly. It may be due to NDF contents and intakes of different diets were similar in this experiment. RSRC diet showed numerically highest in Digestibility of NDF. The ADF digestibility of RSLC, RSGC, RSRC and RSC diets showed significantly different ( $P > 0.05$ ). RSRC diet was found in highest ADF digestibility and lowest in RSLC diet. RSRC diet provided highest CP (9.6%) and highest intake of DM and ADF among all experimental diets. RSRC diet contained more soluble and fermentable protein in the rumen that contributed to highest ammonia concentration in the rumen for efficient microbial activity and it creates efficient ADF digestibility.

#### 4.1.7 Roughages and concentrate ratio and inclusion rate of fodder leaf in total diet

Roughage to concentrate ratio of RSLC, RSGC, RSRC and RSC diets were 49:51, 49:51, 45:55 and 37:63 respectively. All diets except RSC diet used in this study were almost similar (approximately 50:50) in roughage to concentrate ratio. Those diets provided ADG of 412 to 480 g/d and FGR at 5.3 to 6.4 were higher than observation of [20] who found that rice straw was used as a sole source of roughage with concentrate at 50:50 could achieve on Average Daily Gain of 526.9 g/d and FCR at 12 for fattening beef buffaloes. NRC [15] stated that average daily gain is related to the amount and nature of the various metabolites that are absorbed from the digestive tract and these metabolites are determined by the microbial fermentation in the rumen, which in turn is influenced by the proportion of roughage and concentrate in the diet. Fodder leaf proportion to the total diet as DM basis in RSLC and RSGC diet were 21:79 and 27:73 respectively. RSRC diet was 9:91 but RSC diet was 0:100 due to low DM content of Rumex K-1 and absence of fodder supplementation in RSC diet. Preston [17] stated that 20% of DM of total diet in the form of green forage is enough to meet the requirement for the microbial nutrients.

Recommendation made by [6] on the optimum dietary level of fodder tree was 30 to 50% of the total ration on DM basis and that was higher than [17]. Fodder leaf supplementation rate of RSLC and RSGC diet in this study were felt within their recommendations but RSRC diet was found contrary. It has two reasons, first is fodder supplementation rate in this experiment was based on the 1% of live weight as fresh basis and secondly, DM content of Rumex K-1 fodder was very low (6.38%).

4.1.8 Average Daily Gain (ADG) and Feed Gain Ratio (FGR)

Average Daily Gain (ADG) of growing calves fed on all experimental diets was not significantly different. Numerically highest ADG was observed in calves receiving Rumex K-1 supplemented rice straw based diet however, numerically highest Feed Gain Ratio (FGR) was found in *Leucaena* supplemented diet due to its low dry matter intake and moderately high ADG compared to other treatments. This indicated that Rumex K-1 is one of the most suitable protein rich forage to supplement rice straw. The reason may be due to its higher protein content, no tannin content, higher degradability in the rumen compared with either *Leucaena* or *Gliricidia* forage, which results in high fermentable N available in the rumen and it stimulates rumen function. Numerically higher dry matter intake and higher digestibility ( $P < 0.05$ ) of RSRC diet tended to increase overall nutrient intakes of animals, which may provide higher average daily gain of animals. Figure 4 and 5 shows the average daily gain (ADG) and feed gain ratio (FGR) respectively.

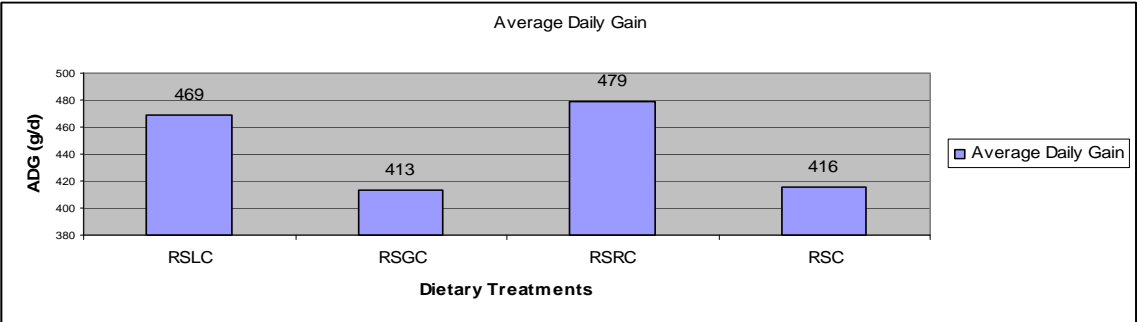


FIG. 4. Average daily gain of 4 dietary treatments

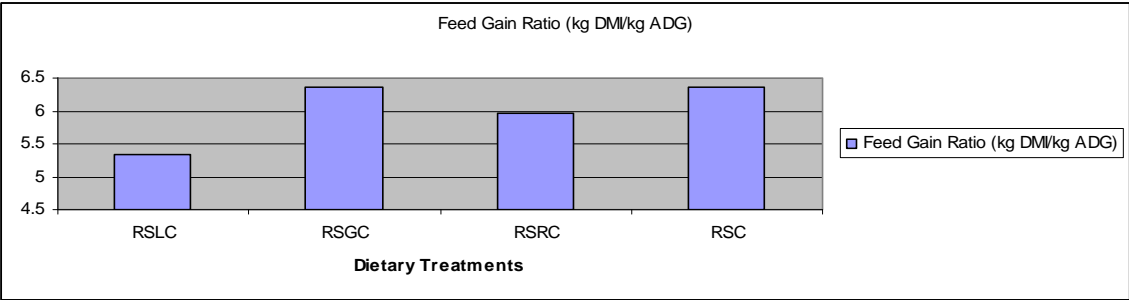


FIG. 5. Feed gain ratio of 4 dietary treatments

## 4.2. Study on the basic indices used for improving conception rate on artificial insemination of dairy cattle at field level

### 4.2.1 Vulva swelling, redness and conception rate

During the oestrus, lips of vulva become in red colour and swelling due to engorgement of blood and glands secrete mucus. Redness of vulva usually persists until end of oestrus and swelling usually subside before end of oestrus. In this study, highest conception rate (83 and 92%) was achieved when animals were inseminated at the period of pronounced vulva swelling and redness. This period was coincided with 12 to 24 hours after onset of oestrus and that period is more favourable for fertilization.

### 4.2.2 Mucus strand, uterine tone and conception rate

Lowest conception rate (70.75%) was found in animals inseminated with little mucus (M1). Little mucus represented that animals are in very early (just come into heat) or later stage of oestrus (beyond 24 hours of oestrus) and [30] also mentioned that early and late oestrus usually show little, thick and milky mucus. It is therefore lowest conception rate related to little mucus may be due to animals were inseminated in early or late stage of oestrus but lowest conception rate indicated here was also felt in acceptable range. Animals showed copious mucus during and soon after standing oestrus (18 hours after onset of oestrus) [32]. Highest conception rate was found in second score of uterine tone and it usually coincides with 12 to 20 hours after onset of oestrus in this study. It was agreed with [27] manual stated that optimum time to inseminate a cow is between 6 to 18 hr after onset of oestrus.

### 4.2.3 Time of insemination and conception rate

Highest conception rate (93%) was found in animals received Artificial Insemination at 12 to 14 hours after onset of oestrus and similar result reported by [29]. The ideal time to inseminate is 12 hrs after the onset of heat and all inseminations carried out within 14 to 24 hours after onset of oestrus have achieved 70% conception rate. Lewis [29] also suggested that good conception rates are achieved in cows inseminated 2 to 26 hours after being observed on heat.

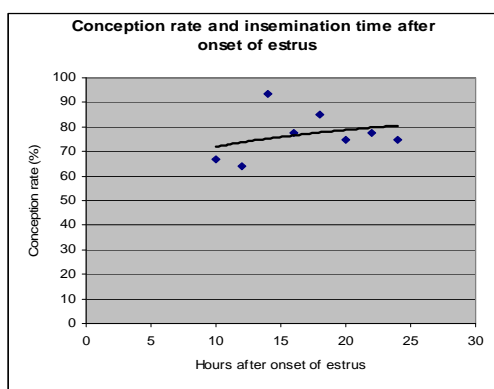


FIG. 6 Conception rate and insemination time after onset of oestrus

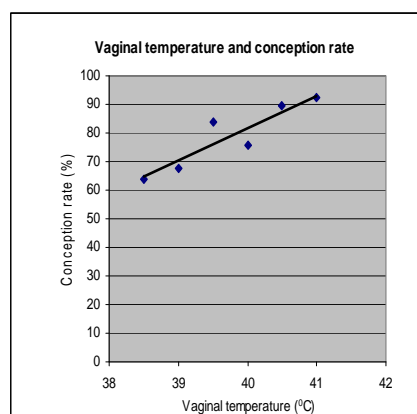


FIG. 7 Conception rate and vaginal temperature

#### 4.2.4 Vaginal temperature and conception rate

It has been reported by various workers that there is a rise in vaginal temperature<sup>e</sup> during the oestrus [34]. Owen [33] cited that elevation in intra-vaginal temperature from 0.3 to 1.1°C may aid in oestrus detection but is not reliable as a sole means of detection, but may have potential, especially in the dairy industry in combination with other detection methods. The efficiency and accuracy in temperature methods were 50 to 74% and 55% respectively.

Highest conception rate (92.3%) was found in cows inseminated at 40.6 to 41°C vaginal temperature and temperature above 39°C to 40.5°C also showed good conception rate (>80%). The possible reason for good conception in this study may be due to the activities of spermatozoa was increased in certain temperature, which is higher than normal body temperature (38 to 38.5°C) and it was agreed with [30]. Incubation media of temperature as high as 41°C causing increase number of spermatozoa to consume their energy reserves due to increase in motility can improve sperm penetration into the ovum thereby enhance fertilization. Finding in this study was also agreed to [26] observation stated that sperm migration in cervical mucus could reflect the penetration capacity of spermatozoa to goat oviduct.

Clapper [25] reported that the onset of a pre-ovulatory temperature increase may be taken as a reliable indicator of the LH surge. The pre-ovulatory surge of LH that occurs in the early hours of oestrus and about 24 hours before follicle rupture is responsible for maturation of oocyte and ovulation [28]. Increasing temperature in oestrus cow's vagina shows that LH concentration is also increasing in the blood and mature ovum will shed from the ovary that will take place less than 24 hours after onset of heat so that chance of fertilization is increased when animals were inseminated during temperature elevated period.

#### 4.3 Improving animal productivity through the use of better selection criteria for offspring from cross- breeding programme

Indigenous cattle are more concentrated in central part of Myanmar and a popular breed is Pyazein, which derives its name from its coat colour – bluish white and origin from white zebu. Pyazein are used as dual-purpose cattle and about 10% of Pyazein cows are used for dairy purpose. Animal health, feeds and feeding and management are important for improved cattle productivity. However, upgrading the genetic potential is also key to improving milk and meat productivity. Improving the genetic potential of cattle including zebus and buffaloes is best achieved through a systematic programme of crossbreeding local cows with high quality, progeny-tested bulls. The operating method used by a specific AI programme will depend, to some extent, on whether semen will continuously be imported continuously or whether it will use locally upgraded bulls. The success of both methods will further depend on the care and feeding of the animals, proper insemination techniques and meticulous record keeping.

In central part of Myanmar and some places, weaning cross- breeding male calves can graze in pasture around the lake or pound for beef purpose. No costing for concentrates and can sell in 16 to 17 months. The prize of the crossbreeding male calf is round about 100 US\$. It is nearly the same benefit with growing up bull calf. Smallholder farmers prefer this managing system for selling as meat type. The rest had already been sold to other townships for the use of natural mating or draught cattle or beef type animals. Although potential bull calves has to be traceable, with frequent exchange of animals among the small farmers, it is very difficult to trace the animal. For the continuation of the selecting bull calves, need to organize medium size farm owners to import top quality frozen semen straws to upgrade the genetic potential.

## 5 CONCLUSION

### **5.1 A comparative study on the effects of different leaf protein supplementation on the performance of pyarzein growing calves fed rice straw based diet.**

The discussion in this experiment defines and highlights the potential use of three types of leaf supplements required for optimal efficiency of utilization of rice straw by cattle with deficient in N or protein. Effects were not different among treatments probably due to short experimental periods (three weeks). Effects of more long-term feeding and higher supplementation rate of leguminous fodders should be explored. Fodder supplementation rate should be increased to 30% of total diet DM to improve N utilization by animals and it could reduce the risk of mimosine toxicity when *Leucaena* is supplemented.

### **5.2 Studies on the basic indices used for improving conception rate of artificial insemination of dairy cattle at field level.**

This study shown that the highest conception rate of 83, 92, 85 and 80%, was found in VS-3 (Pronounced Vulva Swelling), VR-3 (Highest Redness in vulva), M-2 (Thick and hang down mucus) and UT-2 (Firm, tonic and muscular uterus), respectively. In this study, semen factors were minimized by strict quality control of frozen semen straw production whereas inseminator factor were removed by careful examination of oestrus detection and good management practice during AI. Further developments in field-applicable methods to improve conception rate are required through intensive AI training program for field workers and oestrus detection practices for farmers.

### **5.3 Improving animal productivity using better selection criteria for offspring from cross- breeding programme.**

Because dairy bulls are more or less by-products of the dairy industry, the selection of superior bull could only done in medium-size farm where the owner is interested in record keeping and selection. Selection of superior cattle is therefore the responsibility of the breeding station and there must be a permanent set up to keep records and studies. If a calf has a good genetic merit, it should be reared, selected and tested to become a sire if proven good bull.

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# USE OF INDIGENOUS PLANT SPECIES AS TREATMENTS FOR HELMINTHES IN FARM ANIMALS

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## Abstract

Herbal extracts of prairie sagewort (*Artemisia frigida*, family *Asteraceae*), tansy (*Tanacetum vulgare*) and garlic (*Allium sativum*) were infused to test against ovine internal parasites such as *Nematodirus* and *Haemonchus* species in sheep. Twenty-five local Mongolian sheep, infected naturally with *Nematodirus oiratianus* and *Haemonchus contortus*, were selected for the trial. Sheep were allowed to graze on pasture in Taryalan soum (county) of Khuvsgul aimag (province) during the trial period. The efficiency of herbal infusions measured as the percentage reduction of eggs per gram (EPG) of *N.oiratianus* was 13.3%, 15.8% and 11.3% by prairie sagewort, tansy and garlic, respectively. Number of EPG of *H.contortus* was reduced by 22, 20.5 and 17.8% respectively in 5 days later of administration of the infusions. While, infection of sheep with *N.oiratianus* and *H.contortus* from the untreated (control) group were 97.8-100%. The results of the present trial indicated that administration of Prairie sagewort (*Artemisia frigida*, family *Asteraceae*), tansy (*Tanacetum vulgare*) and garlic (*Allium sativum*) infusions to sheep could reduce *N. oiratianus* and *H.contortus* egg shedding 11.3 to 22%.

## 1. INTRODUCTION

Mongolia follows an agrarian economy. The society and the cultural background is mainly nomadic and this play an important role in the future economy of the country [1, 16]. The importance of food security and hygiene is increasing due to escalating demand for foodstuffs by the population. Therefore, it became very vital to organize and intensify control measures against infectious and parasitic diseases in livestock [2, 3]. At present, out of seeding varieties of plants available in Mongolia, 2095 species are herbaceous plants and 348 species are woody and shrubby plants [7]. As recorded, there are 73 species of 36 families plants are used against parasites of animal in Mongolia [2, 4, 9, 18, 19]. More than 170 helminthes species parasitize the livestock in Mongolia [1, 16].

Because of heavy infection with helminthes, animals loose body weight, which leads to dramatic decrease of animal products and often-severe losses due to deaths [1, 2, 3, 8]. Therefore, an alternative means of parasite control is becoming very urgent globally due to high costs associated with the use of commercial anthelmintics and the possibility of developing resistance for the commercial anthelmintics by the parasites [3, 4, 5, 6].

Since the last 15 years, the interest for natural plant products gained a strong impetus because they represent a novel approach to control gastro-intestinal nematodes in animals [3, 5]. Several on-station and field studies have been conducted infected sheep and goats. These studies suggested the presence of bioactive compounds present in these plant species in were able to successfully control the internal anthelmintic parasites and mitigate the burden. Such remedies have been recognized as an economical and sustainable alternative to expensive chemotherapy [3, 5, 6, 7, 8, 9, 12, 13, 14, 15, 18]. Deore (1999) revealed that veterinarians use plants and leaves for the treatment of animals. Garlic has certain effect against lungworms ([www.abdn.ac.uk/organic/organic\\_14c.php](http://www.abdn.ac.uk/organic/organic_14c.php)) and other helminths as well (Wynn, <http://www.altvetmed.com>). The objective of this study was to evaluation of antihelminthic activity of prairie sagewort, tansy and garlic against common helminthes of ruminants.

## 2. MATERIAL AND METHODS

### 2.1 Herbal infusions

Prairie sagewort (*Artemisia frigida*) forage plant commonly grown in prairie-steppe and semi desert zones and tansy (*Tanacetum vulgare*), perennial herb in the compositea family commonly grown in mountain forest-steppe zone in Mongolia were collected from the Centre (Terelj and Nalaikh region of Ulaanbaatar city) and Northern (Tushig county of Selenge province) parts of Mongolia. Air-dried plants samples and garlic cloves (bulbs) were prepared for infusions according to general pharmaceutical procedures [4]. Thirty (30) g of each plant material were ground to powder form and extracted by maceration with 100 ml of boiling distilled water for 2 h. Thereafter the moisture was cool down to 4<sup>0</sup>C and then filtered [2, 4, 19].

### 2.2 Animals and experimental design:

The study was carried out at the small ruminant farm of Taryalan soum county of Khuvsgul aimag province, 450 km from Ulaanbaatar city. The soum is located in mountain forest-steppe of Mongolian vegetation zones predominantly covered with herbaceous plants and the richest diversity of vegetation in Mongolia [4, 19].

Twenty-five local Mongolian sheep averaged weight 15±5.5 and aged between 4-15 months, naturally infected with *Nematodirus oiratianus* and *Haemonchus contortus*. Based on live weights, 25 sheep were allocated into three treatment groups and two control groups (5 sheep to each). Herbal extractants (treatment) were administrated to each group at 100 ml per animal three times daily by 12-hour interval [8, 18].

The treatments are as follows;

T1 – Prairie sagewort

T2 – Tansy

T3 – Garlic

C1 – treated with Almonzol (10% Albendazole suspension, at the rate of 1ml/10 kg of body weight)

C2 – remained as negative control.

Before administration of the infusions and anthelmintic, and thereafter 2, 3 and 5 day post infusion fresh rectal faecal samples were collected and examined microscopically to determine the number of egg per gram (EPG) and presence of *N. oiratianus* and *H. contortus* ova. Counting was done by using flotation fluids (salt and sugar) by McMaster technique ([www.FECPAK.com](http://www.FECPAK.com)).

## 3. RESULTS

### 3.1 Faecal egg count

On the basis of morphological characters of the ova, *Nematodirus oiratianus* and *Haemonchus contortus* were detected in ovine faecal samples (Table I). All sheep were infected with both parasites and 143-152 eggs of *N. oiratianus* and 39-46 eggs of *H. contortus* were detected in a gram of faecal samples. The results suggest that sheep were heavily infected with *N. oiratianus* than *H. contortus*.

TABLE 1. EGG COUNTS OF HELMINTHES AS A RESPONSE TO TREATMENTS

Treatments	Identification of egg types and EPG	
	<i>Nematodirus oiratianus</i>	<i>Haemonchus contortus</i>
T1	143	41
T2	146	39
T3	151	45
C1	149	41
C2	152	46

### 3.2 The reduction of EPG of *N. oiratianus*

Faecal egg count of all sheep checked was lower than the negative control. The efficiency of infusions when expressed as the percentage reduction of EPG of *N. oiratianus* was 13.3% by prairie sagewort, 15.8% – by tansy and 11.3% - by garlic. These values were much lower than Albendazole 10% suspension, which was active against *N. oiratianus* (94%) and *H. contortus* (90.25%).

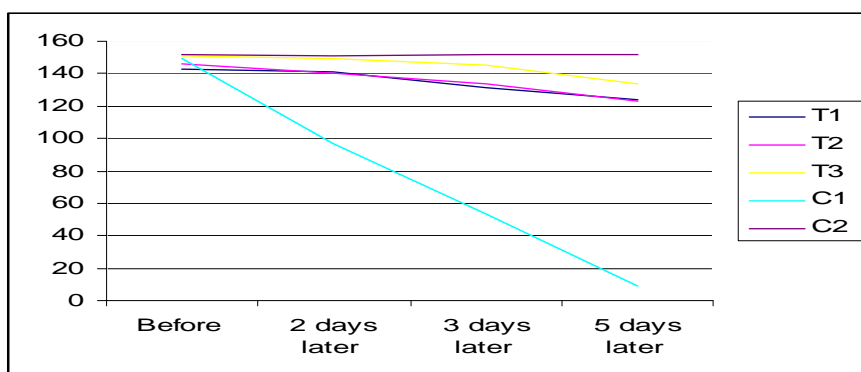


FIG. 1. Mean value of reduction EPG of *N. oiratianus*.

### 3.3 The reduction of EPG of *H. Contortus*

Number of EPG of *H. contortus* was reduced by 22.0, 20.5 and 17.8%, by prairie sagewort, tansy and garlic, respectively in 5 days post infusion. While, infection of sheep with *N. oiratianus* and *H. contortus* from the untreated (control) group were 97.8-100%.

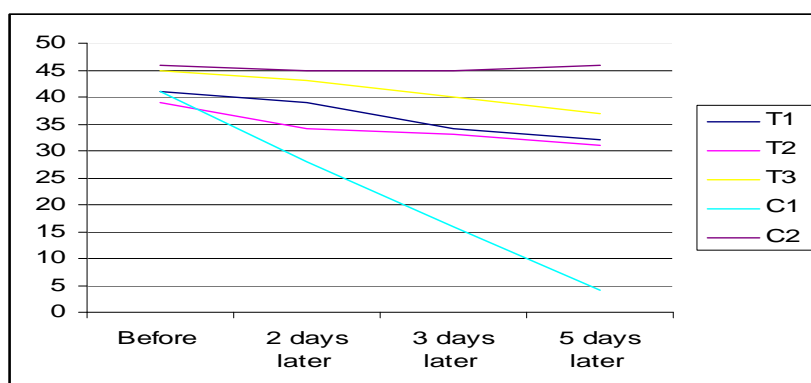


FIG. 2. Mean value of reduction EPG of *H. contortus*.

The results of the present trial indicated that administration of Prairie sagewort (*Artemisia frigida*), tansy (*Tanacetum vulgare*) and garlic (*Allium sativum*) infusions to sheep could reduce *N. oiratianus* and *H. contortus* egg shedding by 11.3-22.0%. There are limited options available for the herbal treatment of helminth infections.

Fresh leaves and bulbs of garlic mixed with molasses and bran for treatment of *Ascaris* and *Enterobios* spp. infection and lungworms (Bastidas, 1969), dried and crushed flower of Wormwood (*Artemisia vulgaris*) - for *Prostrongylus*, *Dictyocaulus* and *Bunostomum* spp. (De Bairacli Levy, 1973), *Artemisia herba-alba* – for caprine *Haemonchus contortus* (Idris et al., 1982) and fresh tansy leaves and flowers (*Tanacetum vulgare*) - for *Nematodirus* spp. (Papchenkov, 1968) were successfully used. Enkhjargal (unpublished data, 1990) found the negative effects of using 10% extract of Prairie sagewort on helminthes eggs, but 30% extract reduced EPG by 25.2%. In this study, we used only 30% herb infusions as well, it is important to conduct further research on preparation of various pharmaceutical types with above plants and investigate and confirm the antihelminthic effects in wider areas. Study on the mode of action of above plant extracts on anthelminthes is important to investigate in the future. .

#### 4. CONCLUSION

Administration of 30% of Prairie sagewort (*Artemisia frigida*), tansy (*Tanacetum vulgare*) and garlic (*Allium sativum*) infusions to sheep reduced *N. oiratianus* and *H. contortus* egg shedding 11.3 to 22.0%.

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# EFFECTS OF MULTI-NUTRIENT FEED SUPPLEMENT IN BEEF CATTLE ON METHANE PRODUCTION, MANURE QUALITY AND RICE YIELD

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## Abstract

The objective of the experiments was to determine the effect of a multi-nutrient feed supplement (MFS) on methane production, and the effect of processing on animal waste to produce compost to improve crop production. Methane is one of the gases that contribute towards global warming and a significant proportion of global methane is produced by rumen fermentation and the breakdown of animal waste. The first of the integrated experimental program carried was the effect of the additional MFS in corn leaf silage with concentrate diet compared to MFS in a low quality of basal diet (rice straw) with a concentrate in beef cattle. Rumen fermentation products including methane gas production were measured along with digestibility, and DLWG. The second phase of the experiment involved the measurement of manures for the nutrient content of the compost and the effect of compost applications on rice yield. The results of the Experiment 1 showed that MFS supplementation increase dry matter digestibility by 15.6%, feed consumption by 7.5%, DLWG by 200% and improve the feed conversion ratio by 187%. The enhanced nutrition improved microbial populations and decreasing methane product from 88.0 to 21.7 mM. The result of the *in vitro* study found that the feed commonly supplied by farmers to their animal could be improved by the use of MFS with methane production decreasing from 42 ml to 25 ml. An *in vivo* experiment using the same rations showed an increase of 5.9, 11.4, 6.6, 4.6, 26.4% in DM consumption, CP consumption, GE consumption, DM digestibility and DLWG, respectively, and improving feed efficiency by 21.3%. N total content of compost derived from control and MFS feeding treatments without and with improved manure management was 168, 156, 150 and 146 g/day. Compost application to paddy rice also increased rice yields 76.5% for dried rice harvesting, and 67.4% for dried rice milling.

## 1. INTRODUCTION

Livestock products, such as meat and milk play an important role in the dietary protein requirements for humans. Animal proteins have been shown to increasing human intelligence, farmer's incomes and reduce unemployment. Increasing rural incomes will have a positive effect on diminishing urban wealth transfer. However, the market demand for animal protein is not being met by current production levels and this has lead to a decrease ruminant population with rising meat consumption.

Beef cattle, buffaloes, goat and sheep populations in Indonesia have decreased by 1.46%, 6.73%, 2.89% and 0.55%, respectively, in recent years [1]. The decrease in cattle population is caused by a long gestation (9-9.5 months), extended calving interval (15-17 months), high calf mortality (7-27%), and cows mortalities of 2.7% [2, 3,4]. These



problems are in part attributed to low feed quality and quantity leading to a reduction in body reserves. Low body reserves are indicated by low body condition scores often leading to delays in first oestrous after parturition and poor milk production [5]. Thus improving the nutrition of cattle will improve condition scores leading to an increase in overall productivity.

It is predicted that meat consumption will increase by 597,700 ton/year requiring the equivalent 3.2 million head increase in the of local beef cattle population by 2010. This increase could be accommodated with 72% being provided from the local beef cattle population [6] and the remainder from imported supplies with 20% from live beef cattle imports and 8% from frozen meat. In addition, national production of milk is only sufficient to meet 25 – 30% of the market; the remainder is made up from imported products. This dependence on imported food products will cause a decline in government foreign exchange reserves. Therefore, the government has put into place a policy to increase food product coming from the livestock sector by increasing livestock productivity, livestock population increases through using locally available resources to increase animal protein supply and to become self sufficient in cattle meat by 2010.

One of approaches being used to increase of cattle productivity is to develop ruminant feeding strategies [7]. This strategy will include appropriate formulations of waste, by-products, and expired of agriculture product and food industries as feed supplement for ruminant animals [8]. However, generally, waste by – products and expired products are poor quality feeds and can only be used as a component in livestock rations [8,9].

Feed formulation, biological evaluation and nutrient analysis using conventional, isotopic and radiation techniques will result in improved feed supplement formulations and development of improved livestock feeding by the use of products such as urea molasses multi-nutrient block (UMMB) and multi-nutrient feed supplement (MFS). Both of these technologies are capable of increasing cattle productivity and improving reproductive performance leading to increased farmers' incomes [8, 10]. These technologies alone can increase farmers' income by up to 20 %, since they are receiving income from the increase in animal productivity alone. Using solely a feeding programme focused on cattle production they did not produce compost, biogas or undertake an improved integration of their crop – livestock production system (CLS). In farms where they had carried out an integrated CLS program the results indicated that additional income can be derived from products such as egg, fish, compost, biogas, etc. Although, they have been undertaking CLS, they have not paid any attention to methane production from the low feed quality of feed consumed or in-adequate manure fertilizer management.

Feed supplements and management systems such as UMMB and MFS increase the supply of nutrients such as carbohydrates, proteins, minerals and vitamins to livestock [11]. If ruminant livestock and rumen microbes are not receiving a balanced diet but have an excess or deficiency in nutrition requirements, the unutilized proportion of the diet will be excreted via urine and faeces, whereas by products from rumen fermentation and manure breakdown such as carbon dioxide and methane will be released as gases. If maintained in manure, N and P make a significant contribution to crop fertilizer management [12, 13]. Methane released from ruminants has a negative impact on both the animal and environment. The CH<sub>4</sub> produced in the rumen is a net loss of dietary feed energy from the host animal, which lowers efficiency of feed utilization [14]. From an environmental perspective, CH<sub>4</sub> released from ruminants contributes significantly to global GHG production [15]. Reductions in methane production can be achieved by using feed supplements, defaunation, chemical inhibitors and biological control [15, 16, 17]. In conclusion, the national demand for animal protein is insufficiently met and is in part

caused by low animal populations, productivity and high consumer demand for animal products (meat and milk), and this is a crucial problem for Indonesian food supplies.

To implementation government policy, the problem of low livestock productivity and supply of animal protein needs to be addressed. A possible solution may be provided from ruminant feeding strategies such as UMMB and MFS to increasing productivity and improve reproductive performance. However, there this not been undertaken as an integrated program for livestock and crop production that measures methane production and determines the socio-economic impact of the changes made to production. An experimental programme of integrated livestock and crop production using feed supplement for ruminants will be undertaken. The program will be carried out in a number of steps. The first step is to study the effect of the addition of a multi-nutrient feed supplement on rumen fermentation product; digestibility and feed conversion in beef cattle. The second step is to measure methane production from dairy cattle and manure management that includes an economic analysis and dissemination of the information to similar regions in Indonesia.

## 2. MATERIAL AND METHOD

The first step is to study the effect of addition of a MFS on rumen fermentation products, digestibility and feed conversion in beef cattle. Two experiments were undertaken: 1) the addition MFS + CLS + concentrate to beef cattle (T1) and 2) the addition of MFS + rice straw + concentrate to beef cattle (T2).

### Experiment 1

A beef cattle feed supplementation experiment using MFS was undertaken. The feeding regimes were T1 with corn leaf silage plus a concentrate and T2 is the T1 feeding regime plus MFS. The composition of concentrate consist of tapioca waste, oil palm cake, oil coconut cake, rice bran, wheat bran, soy sauce waste and mineral mix, where as MFS composition contains molasses, expired date food industries, soy bean meal, urea, *Glyricidae maculata* leaf meal, lime stone, salt and mineral, apart from those, it was the same as concentrate's composition.

Three Ongole cattle (OG) per treatment were used with an average weight of 195 kg. The feed adaptation period was 2 weeks followed by × week for the feeding experiment. Live weight was recorded at the start of the feeding experiment and weekly there after. Feed issue and feed refusals were recorded to determine feed intake. Rumen samples were taken via a stomach tube 4 hours after feeding. It is believed that the activity and population of rumen microbes reach the peak at 4 hours after feeding time [18]. One side of stomach tube was inserted into the rumen through mouth, while another end was connected to the vacuum pump and erlenmeyer. When the pump was on, the rumen liquid was transferred from the rumen into the erlenmeyer. About 100 ml of rumen fluid was collected from each animal. The experiment measured feed consumption, daily live weight gain, and rumen fermentation (ammonia, volatile fatty acid (VFA) total, VFA partials, bacterial and protozoa population and methane gas production.

All of the data collected was tabulated by using Excel® in Microsoft Office 2000 and data was statistically analyzed by using SPSS 11.0.

### Experiment 2

The experiment was carried out at Cimande, Sukabumi (Latitude 6° 55' S, Longitude 106° 55' E), in West Java. The duration of the feeding experiment was three

months plus a 2-week adjustment period prior to the initiation of the nutritional experiment. 15 OG cattle were randomly allocated to two feeding treatments. The first treatment (R1) was a control diet (rice straw *ad libitum*) + 5 kg concentrate/day (farmer mix) and the second treatment was R2 (R1 + 500 g MFS/day). The average body weight for R1 was 249 kg and 235 kg for R2.

Parameters measured including nutrient content of the feed, feed consumption, digestibility, live weight change, and feed conversion. Rumen samples were taken from the rumen of slaughtered cattle and samples were used for an *in vitro* fermentation study [19]. The parameters from the *in vitro* fermentation experiment were gas production, ammonia concentration, volatile fatty acid concentrations, and bacteria and protozoa population estimations. The methodology of those parameters as followed:

### 2.2.1 Total Gas production

Total gas production was measured using a detachable pressure transducer (Bailey and Mackey Ltd, Birmingham, UK). The pressure transducer is used to help in reading the pressure of the headspace gas pressure of each bottle. The transducer had a range of 0 to 15 psi, with an accuracy of  $0.1 \pm 2\%$  at 25 °C. The pressure transducer was connected to the inlet of a disposable Luer-lock 3-way tap (Robinet 3-way tap, France). The first outlet of the 3-way tap was connected to a disposable hypodermic needle (23 gauge x 1.5 inch) while the second outlet was connected to a disposable glass syringe of 60 mL capacity (Sterling Ltd., UK)

### 2.2.2 Ammonia concentration

The NH<sub>3</sub> concentration of the rumen fluid was determined using the Conway technique [20]. This technique uses a glass Conway bowl, boric acid (2 % g/v), sodium hydroxide (40 % g/v), hydrochloric acid (HCl) (0.03 N), Conway indicator (bromo-cressol green : methyl red (1 : 2 wt/wt) in 95 % ethanol solution) and Vaseline.

### 2.2.3 VFA concentration

The concentration of acetate, propionate and butyrate in each sample was determined using Gas Liquid Chromatography (GLC; Hewlett Packard, 3700, USA). The instrument was calibrated using the VFA (acetate, propionate and butyrate) standard for rumen supplied by Supelco (Cat. No 1-1965) Sydney, Australia.

### 2.2.4 Bacteria and protozoa measurement

Bacteria and protozoa population in the rumen was determined by using roll tube method and Methyl green Formaldehyde solution method, respectively [21]. Methane produced during fermentation of the feeds in the culture bottles might be predicted using the equation, based on VFA proportions. The formula's methane (mM) is  $(0.5 \times \text{acetate}) + (0.5 \times \text{butyrate}) - (0.25 \times \text{propionate})$  [22].

Data were analysis using Excel Version 7.0 for Window 95 (Microsoft Corporation, USA) and SPSS Version 7.0 for Window 95 (SPSS Inc. USA) for the statistical estimation of Least Significant Differences (SD). LSD analysis was undertaken when significant differences between treatments were identified using analysis of variance [23].

Manure compost was determined on the collected faeces from each treatment. Faeces from each treatment were maintained in separate piles and each treatment pile was

subdivided. The manure from each feeding treat was subject to either a traditional form of manure management with pile surround by a plank wall to avoid lost of animal waste from experimental site or improved manure management where the pile was cover with small shed. The figure 1 and two describes two treatment of compost processing without and with improved manure management. Animal waste was collected every day during the three months for the feeding experiment. To homogenize the manure for compost processing, the manure was turned over in the pile every three days to ensure the best compost could be attained. Compost processing was completed within 14 days, and the compost stored in the respective composite treatments piles until applied on the rice paddy fields as fertilizer.

The climate around the location of experiment has an average rainfall of about 4470 mm with a frequency of 191.2 rainy days per year (minimum, 4009 mm with frequency 178 rainy days per year and maximum 5407 mm with frequency 213 rainy days per year). Average temperature was 23.3 – 24.5 ° C, humidity 78 – 86 % and topography 190 – 350 m above sea level.



*FIG. 1. Animal waste collection structures without management system*

At the end of the animal manure collection period each manure pile weight was measured and prepared for fertilizing there respective rice plots. The manure composts were sub-sampled for dry matter and N content determination by oven drying at 60 °C and Kjeldahl N method, respectively. Compost application were used as fertilizers of Mira I variety rice produced by mutation breeding at National Nuclear Energy Agency [24].



*FIG. 2. Animal waste collection structures with improved management systems*

The irrigation system used was tertiary and was separated by each treatment for entry and exit of water supply, so that the fertilizer treatment was not influenced by external factors.

The rice growth experiment was carried out using 8, 15 x 3.5 m<sup>2</sup>/plots in the low lands (latitude, longitude). Paddy manure fertilizer regimes were divided on two treatments blocks with two replicates for each manure management system. The treatments plots are shown in Table I.

TABLE I. RICE MANURING TREATMENTS APPLIED TO MIRA I VARIETY OF PADDY RICE

Improved management		Traditional	
2 A	1 B	1A	2 A
1 A	2 B	2 A	1B

Notes: 1: Control feed; 2: MFS treatment; A: Replication of 1 and B replication of 2; Compost fertilizer/organic fertilizer application rate was 5 ton/ha, and an organic fertilizer/ha used at the rate of 200 kg Urea + 100 kg Super phosphate – 36 (SP-36) + 100 kg/KCl.

### 3. RESULT

#### 3.1. Experiment 1

MFS feed intervention in beef cattle were undertaken, and the proximate analysis for the dietary ingredients is shown in Table II. The crude protein (CP) content of MFS was higher than corn leaf silage (CLS) and concentrate being 21.34, 11.9 and 8.6% respectively. The composition of concentrate and MFS was different. It is presented on Table III.

TABLE II. DRY MATTER (DM), CRUDE PROTEIN, CRUDE FIBRE AND GROSS ENERGY CONTENT OF TWO EXPERIMENTAL FEED

Materials	DM (%)	CP (%)	CF (%)	GE (Kcal/kg)
Corn leaf silage (CLS)	20.6	11.9	11.5	1.08
Concentrate	88.7	8.6	12.7	3.17
MFS	88.5	21.3	245	3.21

TABLE III. INGREDIENTS COMPOSITION OF THE CONCENTRATE AND MFS

Concentrate			MFS	
No.	Feed materials	(%)	Feed materials	(%)
1	Tapioca waste	60	Tapioca waste	9
2	Oil palm cake	2	Molasses	10
3	Oil coconut cake	2	Expired date food industries	30
4	Rice bran	18	Wheat bran	12
5	Wheat bran	8	Soy bean meal	2
6	Soy sauce waste	8	Rice bran	14
7	Mineral mix	2	Urea	2
8	-	-	<i>Glyricidae maculata</i> leaf meal	2.5
9	-	-	Soy sauce waste	15
10	-	-	Lime stone	1
11	-	-	Salt	1
12	-	-	Lactation mineral	1.5

Rumen fluid parameters measured in the experiment are shown in Table IV. MFS feed to beef cattle significantly increased bacterial and protozoa count in rumen by 53 and 22% respectively. Degradable N contents in MFS increased rumen ammonia concentration to 243.7 mg/L and that was significantly higher than of control fed cattle at 195.5 mg/L. MFS treatment significantly increased propionate concentration compared to that found in the control group. The estimated methane production from the cattle feed MFS was significantly reduced from 88.0 to 21.7 mM.

TABLE IV. THE EFFECT OF MFS FEED TREATMENT ON A RESULT OF FEED FERMENTATION BY MICROBIAL RUMEN

Parameters	T1 (control)	T2 (supplemente d)	P value	SE
VFA <sup>1</sup> partial (mM/ml)				
Acetate	170.0	49.7	0.030	29.7
Propionate	10.7	26.6	0.023	5.7
Butyrate	12.9	8.3	0.088	3.9
VFA total (mM/ml)	193.6	84.6	0.011	28.7
NH <sub>3</sub> (mg/L)	195.5	243.7	0.001	20.3
Bacterial total count (colony/ml)	2.02 x 10 <sup>9</sup>	3.09 x 10 <sup>9</sup>	0.006	0.49 x 10 <sup>9</sup>
Protozoa total count (cell/ml)	1.75 x 10 <sup>5</sup>	2.13 x 10 <sup>5</sup>	0.001	0.16 x 10 <sup>5</sup>
Gas total (ml)	224	219	0.010	2
Methane (mM)	88.0	21.7	0.043	16.5

<sup>1</sup>VFA = volatile fatty acid

The effect of MFS supplementation on dry matter digestibility, feed consumption, daily live weight gain and feed conversion ratio is shown in Table V. Dry matter

digestibility was significantly increased by 15.6% when compared to that of the control. Likewise live weight gain and feed conversion were significantly improved with MFS.

TABLE V. FEED CONSUMPTION, DRY MATTER DIGESTIBILITY, DAILY LIVE WEIGHT GAIN (DLWG) OF OG CATTLE AND FEED CONVERSION RATIO (FCR) FOR CONTROL AND MFS INTERVENTION

Parameters	T1 (control)	T2 (supplemented)	P value	SE
Feed Consumption (g)	5880	6323	0.046	108
Dry matter digestibility (%)	60.8	70.0	0.001	2.4
DLWG (g/d)	129	398	0.014	70
FCR (g DM/g DLWG)	45.6	15.9	0.030	8.8

### 3.2. Experiment 2

#### 3.2.1. *In vitro* study

The results of *in vitro* fermentation of the dietary components are shown in Tables VI and VII. The substrates used in the *in vitro* studies were rice straw (RS), concentrate, MFS and the two fully formulated feed treatments.

The control feed (RS+C) produced more gas (167 ml) in a 48 h incubation period than the control + MFS diet (104 ml). Similar differences were seen in methane production where the control diet produced 42 ml in 48 h and the MFS treatment produced 25 ml (Table VI).

TABLE VI. THE RESULTS OF *IN VITRO* STUDY ON EXPERIMENTAL FEED RELATED TO METHANE CONCENTRATION

Experimental feed	Gas total (ml)		Methane Gas (ml)		Methane (% total gas)	
	24 h	48 h	24 h	48 h	24 h	48 h
Rice straw (RS)	43	91	15	19	34	20
Farmer's concentrate (FC)	47	62	13	15	28	24
MFS	103	123	31	34	30	28
RS + C	132	167	36	42	27	25
RS + C + MFS	54	104	12	25	22	24

Dry and organic matter digestibility, bacteria and protozoa count and ammonia concentration were changed by MFS supplementation (Table VII).

TABLE VII. THE RESULTS OF *IN VITRO* FERMENTATION STUDY OF EXPERIMENTAL FEEDS

	pH	DDM (%)	DOM (%)	Total Bacteria count (x10 <sup>9</sup> colony/mL)	Total Protozoa count (x 10 <sup>6</sup> cell/mL)	Ammonia (mg/L)
Rice straw (RS)	6.59	43.9	47.2	14.0	24	157
Concentrate (C)	6.49	62.7	65.1	2.4	6	168
MFS	6.43	66.9	72.1	8.0	12	275
RS + C	6.66	61.6	64.5	6.0	9	164
RS + C + MFS	6.49	65.7	65.4	17.6	20	194

### 3.2.2. *In vivo* study

Feed composition of concentrate in Experiment II was different with Experiment I, it was shown on Table VIII. It was supported the result of crude protein content. The proximate analysis of the diets used in the *in vivo* experiment is shown in Table IX. The crude protein contents of the basal rice straw and concentrated were 5.0% and 12.2%, respectively.

TABLE VIII. FORMULA'S COMPOSITION OF CONCENTRATE AND MFS

Concentrate			MFS	
No.	Feed materials	(%)	Feed materials	(%)
1	Tapioca waste	35	Tapioca waste	9
2	Corn	14	Molasses	10
3	Fish meal	2	Expired date food industries	30
4	Oil palm cake	4	White bran	12
5	Mineral mix	2	Soy bean meal	2
6	Rice bran	20	Rice bran	14
7	Oil coconut cake	4	Urea	2
8	Soya bean meal	1	<i>Glyricidae maculata</i> leaf meal	2.5
9	Soy sauce waste	3	Soy sauce waste	15
10	Wheat bran	15	Lime stone	1
11	-	-	Salt	1
12	-	-	Lactation mineral	1.5

TABLE IX. CONTENT OF DRY MATTER (DM), CRUDE PROTEIN (CP), ENERGY, ASH, NDF, ADF, CA AND P OF THE EXPERIMENTAL FEEDS

Feed materials	DM (%)	CP (%)	Energy (Kcal/kg)	Ash (%)	NDF (%)	ADF (%)	Ca (%)	P (%)
Rice straw	92.1	5.0	3346	16.9	68.4	40.5	0.19	0.11
Farmer's concentrate	94.2	12.2	4061	12.7	43.6	29.9	0.19	0.80
MFS	91.3	17.7	4145	10.5	18.9	12.8	1.65	0.54

DM, CP, and GE intakes along with DM digestibility are shown in Table X.

TABLE X. DRY MATTER (DM) INTAKE, CRUDE PROTEIN (CP) INTAKE, GROSS ENERGY (GE) INTAKE, AND DRY MATTER (DM) DIGESTIBILITY FOR THE NUTRITIONAL TREATMENTS

Parameters	Control	MFS	P
DM intake (g/day)	11848 <sup>b</sup> ± 166	12551 <sup>a</sup> ± 32	0.004
CP intake (g/day)	881 <sup>c</sup> ± 11	981 <sup>a</sup> ± 2	0.000
GE intake (Kcal/day)	42477 <sup>b</sup> ± 587	45258 <sup>a</sup> ± 107	0.002
DM digestibility (%)	75.4 <sup>b</sup> ± 1.2	78.8 <sup>a</sup> ± 0.8	0.048

Daily feed consumption, live weight gain and feed conversion for the nutritional regimes are shown in Table XI.



TABLE XI. FEED CONSUMPTION, DAILY LIVE WEIGHT GAIN AND FEED CONVERSION FOR THE TWO NUTRITION TREATMENTS IN ONGOLE CATTLE

Parameters	Control	MFS	P
Feed consumption (g DM/d)	11848 ± 166	12551 ± 32	0.004
Daily weight gain (kg/d)	0.821 ± 0.14	1.038 ± 0.21	0.052
Feed conversion ratio	17.0 ± 3.0	14.0 ± 2.3	0.081

The results for individual and total VFA concentrations are shown in Table XII. Feeding MFS increased propionate concentrations leading to a decline in predicted methane production (Table XIII).

TABLE XII. THE EFFECT OF FEEDING MFS ON INDIVIDUAL AND TOTAL VOLATILE FATTY ACID CONCENTRATIONS IN RUMEN LIQUID OF ONGOLE CATTLE

Parameter	Control	MFS
Acetate	188.8	155.2
Propionate	33.1	37.2
Butyrate	20.3	28.9
Total	242.2	221.4

TABLE XIII. ESTIMATED METHANE CONCENTRATION IN RUMEN LIQUID OF ONGOLE CATTLE

Treatment	Methane (mM)	Methane decrease (%)
Control	94.2	
MFS	80.2	14.86

### 3.2.3. Fertilizer management

The proximate analysis of composted manure during the experiment is shown in Table XIV. The total N content of the composted manure from each composting box are 168 and 150 g/day for no management group, whereas for the group of applying improved management are 156 and 146 g/day for control and MFS sourced manures, respectively.

TABEL XIV. DRY MATTER, NITROGEN CONTENT AND THE AMOUNT OF FEACES EXCRETED PER DAY FROM EACH GROUP

Parameters	Control		MFS	
	- Management	+ Management	- Management	+ Management
N content (%)	11.6	10.8	11.3	11.0
Total amount of faeces (g)	5993	5993	5379	5379
Faecal DM (%)	24.1	24.1	24.6	24.6
Total DM of faeces (g)	1446	1446	1323	1323
Total N (g)	168	156	150	146
Amount of N applied for each plot of rice growth experiment (8.15 x 3.5 m <sup>2</sup> ) (g)	3.4	3.1	3	2.9

Good manure compost management for both control and MFS feed treatments improved the yield of dried rice harvesting by 76.5% or 67.4% for milled dried rice (Table XV).

TABLE XV. EFFECT OF FEED INTERVENTION AND MANURE MANAGEMENT ON RICE YIELD

Yield			Feed treatments			
			Control		MFS	
			- Management	+ Management	- Management	+Management
Harvested	dried	rice	1.81± 0.94			
(tons/ha)				2.19 ± 0.94	1.62 ± 0.94	2.86 ± 0.27
Milled dried rice (tons/ha)			1.07 ± 0.58	1.28 ± 0.47	0.89 ± 0.58	1.49 ± 0.11

#### 4. DISCUSSION

##### 4.1. Experiment 1

To achieve a live weight gain of 0.75 kg/day beef cattle require 622 g CP/day [25] and this level of CP intake was exceeded by the MFS feeding regime (646 g CP/day) but the cattle were unable to achieve 0.75 kg/day live weight gain indicating an on going energy deficiency that has not been met at this level of protein intake. The control diet provided 553 g/day of crude protein suggesting a major energy deficiency with probably adequate protein intakes to achieve a growth rate of 129 g/day.

MSF feeding improved rumen fermentation as indicated by an increasing of microbial population by 53%, dry matter digestibility by 15.6% and feed consumption by 7.53%. MFS thus contains essential nutrient and maintains fermentation equilibrium for the available degradable N and carbohydrate to maximise microbial growth. Essential minerals for ruminant grow include sulphur, copper and zinc [26] and these minerals have been measured previously in MFS [27]. These minerals play an important role on rumen fermentation, because S and Zn increase microbial activity and protein synthesis [28] and Cu improves crude fibre digestibility [28] microbes in rumen.

Degradable N content of MFS is derived from urea and *Glyricidia maculata* meal that increases rumen ammonia concentrations to 244 mg/L and higher than the control diet at 196 mg/L. Preston and Leng [7] reported that 50 - 250 mg/l ammonia concentration content is required to maximize rumen fermentation but increasing levels of N intake beyond those to reach these levels of rumen ammonia stimulated intake and improved live weight responses. MFS supplementation has a positive effect on rumen fermentation with propionate concentration increasing, and this elevation results a predicted decline in methane product from the rumen. Methane product in the rumen is influenced by animal type, feed quantity and quality [29]. Decreasing methane gas product has a positive affect on energy utilization with increasing DLWG with a more efficient use of feed.

Kurihara *et al.*, [30] reported that ruminants with lower methane production had a higher DLWG than those with higher rates of methane production. In addition, Kurihara *et al.*, [31] also reported that feed interventions influenced DM consumption, DLWG and methane production. Joblin, and Nolet *et al.*, [32,33] calculated that if methane production was fully converted to acetate, propionate and others fermentation product, the additional energy would increase DLWG by an addition 4 – 15 %.

Less methane gas production in the rumen of animal fed MFS indicates that feeding with MFS becomes an option for reducing methane release from ruminants with a lowering of air pollution/greenhouse effects and higher ruminant productivity.

## **4.2. Experiment 2**

### *4.2.1. In vitro study*

Blummel [34] reported that the products of rumen fermentation are volatile fatty acids, microbial protein and gasses. Gas production is used as an indirect measurement of microbial activity from feed digestion with associated products of short chain fatty acids and microbial biomass. Theodorou *et al.*, [35] reported that the gas production technique is capable of evaluating rumen fermentation outcomes of feed samples. To achieve accuracy of feed evaluation using the gas production technique the gases need to be partitioned to determine the relationship between cumulative gases produced and the amount of feed fermented [36]. Total gas and methane production were reduced by MFS feeding with gas total production dropping from 167 ml to 104 ml and methane production from 42 ml to 25 ml. However, other researchers have suggested that data resulting from *in vitro* study should be supported *in vivo* studies [37].

### *4.2.2. In vivo feeding experiment*

Dry matter digestibility of feed supplied with MFS was higher than control feed. The *in vitro* experiment indicated lower total gas and methane production, and in the *in vivo* experiment, this improvement in fermentation resulted in a DLWG increase of 26.4% and a feed efficiency improved 21.3%. Feeding with MFS to improve feed quality with an increased DLWG of 1.2 kg/d compared with cattle consuming the medium quality control feed that achieved DLWG of 0.5 kg. This change in DLWG is consistent with other reports that have successfully reduced methane production [31].

Whitelaw *et al.*, [38] reported that methane production was negatively correlated with propionate proportion in the rumen resulting from feed fermentation. Thus, if the propionate proportion was raised then methane released as lost energy to the air will be lowered. In this feeding experiment propionate acid concentration was 37.24 mM with MFS in the diet and higher than that for control fed cattle at 33.1 mM. This resulted in a predicted methane production of 80.2 vs. 94.2, respectively or a decrease of 14.9% in methane output.

Improvement of FCR due to MFS supplementation was a consequence of producing a lower methane output. Kurihara, *et al.*, [30] reported that the lower the methane production, the higher of daily weight gain. McCrabb and Hunter [39] also reported an improvement of 0.1 kg/day in DLWG for cattle with a 13% reduction in methane production when non-treated control cattle had a DLWG of 0.5 kg/day.

### *4.2.3. Fertilizer management*

Lost N in the faeces seemed to be higher when the cattle were fed the control diet compared to MFS treatment regardless of the manure management system imposed. This indicates that the increased N supplied to the cattle was utilized to increasing productivity. Compost derived from improved manure management was improved in quality as indicated by increasing rice yields by 76.5%/ha dried rice harvesting or 67.4%/ha/dried rice millings.

According to published data for Mira I rice variety the average yield and potency of yield was 6.29 and 9.2 ton/ha dried rice milling [24]. The decrease in yield of this experiment was probably due pest attack and diseases during compost application. Despite the crop health problems, rice yield still increased 76.54%/ha dried rice harvesting or 67.42%/ha/dried rice milling and assembly reflects the differences in compost processing achieved with better manure management. However, further research is required to manage compost applications to ensure that there are no pest or disease problems associated with applications of compost to maximize rice yields. Mira I variety has been widely disseminated and is now planted in 1 million hectares across 23 of 33 Indonesia provinces.

## 5. CONCLUSION

By multi-nutrient feed supplementation (MFS) to beef cattle fed either medium quality of basal diet and low quality concentrate or low quality of basal diet with a medium quality concentrate found that MFS was able to increase dry matter digestibility, feed consumption, daily live weight gain and feed efficiency through decreased methane production and decreased N loss in the faeces.

A better manure management for the composting process to produce a high quality compost increased rice yield of Mira I, both on a per ton dried rice harvesting/ha or dried rice milling basis, although pest and diseases had attacked the paddy rice during compost application.

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# INTEGRATED APPROACHES FOR IMPROVING LIVESTOCK PRODUCTION USING INDIGENOUS RESOURCES AND CONSERVING THE ENVIRONMENT ON SMALLHOLDER FARMS IN NORTH-EAST THAILAND

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## Abstract

Two experiments were undertaken to investigate approaches to reducing ruminant methane production, and N and P losses, on small-scale dairy farms in northeast Thailand. Cassava hay (CH) supplementation and three methods of manure management (non-covered, covered, covered with EM-treated) were evaluated. Methane production was predicted using rumen volatile fatty acid concentrations, while N and P contents in urine, fecal and fertilizer were also measured. Supplementation with CH tended to decrease methane production by changing the proportion of volatile fatty acids. Milk yield and compositions were the same for non- and supplemented groups, however, milk fat content increased with CH supplementation. Nitrogen and P excretions were not decreased by CH supplementation. Manure management strategies (covered and covered with EM treated) did not reduce N and P losses during preservation when compared with non-covered group. EM-treated, covered-manure decreased N losses but not the P content of fertilizer when compared with the covered treatment. Based on these results it could be recommended that CH supplementation was advantageous to smallholder-dairy farms and manure preserving with covering should be given strategic consideration.

## 1. INTRODUCTION

Cassava or tapioca is an annual crop grown widely in tropical and sub-tropical areas, particularly in Thailand. Cassava thrives in sandy-loam soils with low organic matter and low rainfall regions [1, 2]. It has been found that cassava hay harvested at an early stage of growth (3-4 months) contains up to 25 % crude protein (CP) and with a good amino acids profile. Condensed tannins (CT) and hydrocyanic acid (HCN) concentrations were low in both cassava leaves and cassava hay (CH). Sun drying significantly reduced HCN concentrations [4, 5]. Condensed tannin content between 2-4 % of DM can help protect protein from rumen digestion, thereby increasing the amount of rumen by-pass protein. In the presences of CT, proteins can form tannin-protein complexes (TPC) in the rumen that will dissociate in the lower gut for digestion and absorption [4, 6]. If slow release PEG is supplied with a high CP content diet then there is higher amount of microbial protein available post ruminally because of better efficiency of microbial protein synthesis [7].

Methane (CH<sub>4</sub>) is one of the GHG contributing to global warming [8, 9]. Global emission of methane from livestock, particularly ruminants, is the largest single source of global anthropogenic methane production [8]. Methane production from livestock in Thailand is predicted to increase at approximately 4.6 % per annum over the next three decades with the increasing livestock population. There have been many strategies proposed to reduce methane production from ruminants and these have been comprehensively reviewed by [10, 11]. Research has found that tannin containing feeds leads to a protein-sparing effect in the rumen with reductions in methane production and



N excretion to the environment, thereby reducing emissions of environmental pollutants while increasing meat, milk and wool production. Feeding strategies need to be designed to exploit these beneficial effects of tannins [12]. Higher molar proportions of propionate in *in vitro* fermentation and lower protozoal counts have been produced by tannins in a RUSSETIC evaluation [13] and this was consistent with higher efficiency of microbial protein synthesis that has been observed in presence of tannins. Although effects, *in vivo*, of tannins on rumen protozoal counts and molar proportions of short-chain fatty acids are inconsistent [14], some reports show lower protozoal numbers [15] and a higher molar proportion of propionate [16] in presence of tannins. Current experiments feeding whole cassava crop (cassava hay) containing CT supplementation to ruminants shifted rumen microorganisms populations particularly lowering rumen protozoal populations [17, 18]. In addition, research by [19] showed that supplementation of ground mango steen peel containing high level of CT and saponins resulted in decreased rumen protozoal populations. The changes in population of protozoa and/or propionic producing bacteria are predicted to effect methane production.

However, due to the lack of research on reduction of methane production in ruminants fed on tropical feeds our experiment aims to study the reduction of methane production in ruminant using nutritional strategies in a tropical environment. Utilization of local feed resources such as cassava hay that contain natural CT could be introduced into feeding systems practiced by farmers to both enhance productivity and reduce methane production to environment.

Cow manure is an important product of dairy farms, both in terms of smallholder farm income and as a problem for waste management. Cow manure is generally removed from the barns every day and stored nearby, where it is spread over the soil surface in an open area and sun-dried. Dry manure is commonly used on their farms or sold to farmers in other locations, which is used as a fertilizer for forage plots, vegetables, orchard trees, etc. Chemical analyses of wet cow manure for N, P and K found that the levels ranged from 0.96 to 2.12%, 0.33 to 0.79%, and 0.53 to 0.87%, respectively [20]. Although each farm may have only small numbers of dairy animals, when a large number of farms exist in a small area, the bulk of animal wastes produced each day can create long-term environmental problems for farmers themselves, and for other people in neighbouring areas. Liquid wastes from dairy farms can contaminate water resources and public waterways. Piling and drying of manure on land surfaces without vegetation can result in leaching and seeping of inorganic and organic matter into underground water.

Many dairy farms, particularly in smallholdings, manage farmyard manure in a very simple way often leading to N volatilization. Livestock manure is stored near milking and/or animal houses, sun-dried and applied to crop fields once or twice a year is the traditional practice of most farmers. Therefore, the strategies of manure management, which are simple and low cost, should be recommended to farmers. At present, the technology of waste degradation using microorganisms has not been widely adopted in Thailand and would therefore be an alternative strategy for improving the efficiency of manure management before manure applications to crops as a fertilizer.

## 2. MATERIAL AND METHODS

### 2.1. Experiment I. Feeding strategies to reduce ruminal CH<sub>4</sub> production and improve milk productivity

Seventeen smallholder-dairy farms from two sites (Nampong (latitude 16° 46'N Longitude 101° 42'E) and Kranuan (Latitude 16° 42'N Longitude 103° 5'E), Khon Kaen Province) were selected for on-farm evaluation and as demonstration farms. two lactating crossbred Holstein cows on each farm (34 heads) were allocated to one of two groups (as follows): Group I - existing feeding system (control) and Group II - existing system supplemented with 2 kg/h/d cassava hay. In the existing feeding system (Group I) all cows received roughage *ad libitum* while concentrate was provided in proportion to daily to milk yield (1 litres:2 kg concentrate). Average of the ingredients and chemical compositions is shown in Table I. Roughage and concentrate samples were taken from each farm week for chemical analyses. Individual intakes of roughage and concentrate, and daily individual milk yields were recorded for 60-days. Milk samples were collected twice a month for fat, protein, lactose, total solids, and solids-not-fat concentration determination using Milkoscan (Model 133 V3 7 GB). Total manure excretions were recorded daily, and manure samples were taken every week for chemical composition analysis that included dry matter, ash, N, and P [21]. Rumen fluid was sampled monthly using a stomach tube aided by a vacuum pump. Ruminant pH and temperature were measured immediately (HANNA instruments, HI 8424 microcomputer) and volatile fatty acids concentration were determined by high performance liquid chromatography (HPLC model Water 600; UV Detector, Millipore Corp.) according to the method of [22]. Methane production was estimated using ruminal volatile fatty acid concentrations as recommended by [23].

TABLE I. AVERAGE INGREDIENTS AND CHEMICAL COMPOSITION OF CONCENTRATE

Items	Proportion, %DM	Standard deviation
Ingredients		
Cassava chip	63.4	2.33
Rice bran (Fined)	10.2	1.26
Soybean meal (44%)	14.7	2.54
Brewers' grain	16.2	1.98
Palm kernel meal	8.0	1.03
Coconut meal	6.7	1.11
Urea (46%N)	1.4	0.26
Molasses	2.1	0.21
Sulphur	0.3	0.10
Premix minerals (Dairy)	0.9	0.07
Chemical composition		
DM (%)	91.2	1.22
CP (%DM)	17.8	1.36
TDN (%DM)	72.1	2.64
Price (Baht)	4.3	0.47

### 2.1.2. Experiment II. Manure management strategies to reduce N and P loss to environment

Comparison of three methods of manure management was conducted on a demonstration farm as follows: A (Control) - general method practiced by farmers, without roofing and coverage, and without technology of using effective micro organisms, B (Intervention 1) - with roofing and coverage to protect from sun light and wind and C (Intervention 2) - intervention method 2 plus use of effective micro organisms (EM) to enhance degradation of manure's organic matters (could you please provide details of EM).

Randomized complete block design (RCBD) were used in this experiment. Thirty three crossbred Holstein cows were held in a covered area with cement flooring to determine feed intake and total excreted manure for a 7-days period (3, 7-day periods in three months). Samples of manure from each 7 days were immediately taken for analyzed of N and P excretion. Excreted manures of each cow from each farm were sampled for analysis of DM, and N and P to measure the reduction of N and P loss to environment. The remaining N and P in manures were maintained for capturing N and P efficiency comparison. The methods used in the analysis were according to [20].

Three plots of cassava and legume (intercropping) were prepared for manure applications. The soil was ploughed, ridged and each plot sub-divided into three sub-plots (4 x 8 m each). Cassava stakes cut into 15-20 cm lengths from healthy and diseases free stems of the variety Rayong 72 were used. A row spacing of 40x60 cm (between stem 40 cm. and between row 60 cm.) was used in all plots. Legume (*Thapra stylo*; *Stylosanthes guianensis*) was planted every 2 rows per 4 rows of cassava with the same spacing. Weeding was undertaken on all the plots twice, 1 and 2 months after planting. Fertilizer from the three methods of manure management above was applied 4 month after planting at the rate of 2 tons/ha. The CS relied on rainfall for growth and the annual rainfall was 127 mm. Crop products including cassava and legume hay were harvested four times (at manure application time and every three months after application) and cassava root-crop yield was determined one year of planting. Crop products were used for home-consumption, sold for income or used as on-farm animal feeds.

All data were subjected to Analysis of variance using Proc. GLM [24]. Treatment means were statistically compared using Duncan's New Multiple Range Test.

## 3. RESULTS AND DISCLUSIONS

Table I shows ingredients and chemical composition of the concentrate. The two main energy sources were cassava chips and rice bran while protein sources were more variable. The ingredients used depended on availability of feeds at the sites and the price of ingredients. Moreover, price of the farmer produced concentrate was lower than commercial feeds which ranged from 6-8 Baht/kg (1 US\$ = 35 Baht).

Voluntary feed intakes, N and P excretion, and dry matter (DM) digestibility are shown in Table II. Total concentrate and roughage intakes of dairy cows were not different across treatments. However, CH supplementation tended to reduce the amount of concentrate feed used in all periods ( $P = 0.07$ ). This implies CH supplementation can reduce the cost of milk production in smallholder farmers. This is consistent with the findings of [4] Wanapat et al., (2000) who suggested that CH supplementation significantly reduce concentrate use to a lower ratio of 1:4 (concentrate: milk). Nitrogen intake was higher in CH supplemented group ( $P < 0.05$ ) while P intake was not different. CH supplementation significantly increased N excretion via faeces ( $P < 0.05$ ) reflecting a

higher N intake but as a percentage of N intake there was not difference between the nutritional regimes in N excretion. This result may be due to N supply from CH supplementation being in balance with the improved energy intake. Moreover, P intake or excretion was not affected by CH supplementation. However, [4] found P content of CH was 0.22% DM and this is considered to be below that required for normal growth rates [25]. Dry matter digestibility was not different between the groups.

TABLE II. EFFECT OF CASSAVA HAY (CH) SUPPLEMENTATION ON VOLUNTARY FEED INTAKE, NITROGEN AND P EXCRETION, AND DRY MATTER DIGESTIBILITY

Items	Control	CH supplementation	SEM
Voluntary feed intake, kg/h/d			
Total	12.9	12.6	0.42
Roughage	7.2	7.3	0.79
Concentrate	5.7	5.3	0.66
Faecal output, kg/h/d	6.6	7.5	1.46
Nitrogen intake, g/h/d <sup>1</sup>	288.7 <sup>a</sup>	328.9 <sup>b</sup>	4.23
Phosphorus intake, g/h/d	107.1	109.8	3.48
Nitrogen excretion, g/d	138.9 <sup>a</sup>	154.2 <sup>b</sup>	5.32
Nitrogen excretion, % of N intake	48.1	46.9	1.84
Phosphorus excretion, g/d	38.0	40.2	2.13
Phosphorus excretion, % of P intake	35.5	36.6	1.32
Dry matter digestibility (%)	49.3	49.2	2.94

<sup>1</sup> Nitrogen and P excretion included those from CH supplement.

Milk yield and milk composition for the feeding treatments is shown in Table III. Milk yields were similar for the feeding treatments. Milk composition including fat, protein, lactose, total solids and solids-not fat percentages were also the same for the feeding groups. However, milk fat and solids-not fat tended to be higher in the cassava hay supplemented group ( $P < 0.09$ ).

TABLE III. EFFECT OF CASSAVA HAY SUPPLEMENTATION (CH) ON MILK YIELD AND COMPOSITION

Items	Control	CH supplementation	SEM
Milk yield (kg/h/d)	12.9	12.7	0.33
Milk composition, %			
Fat <sup>a</sup>	3.6	3.7	0.28
Protein	3.0	3.1	0.31
Lactose	4.9	5.0	0.29
Total solids	12.5	12.1	1.02
Solids-not fat <sup>a</sup>	8.7	8.8	0.27

<sup>a</sup>  $P < 0.09$

The effect of cassava supplementation on rumen contents is shown in Table IV. Ruminal pH, temperature and total volatile fatty acids were not different between treatments. Ruminal temperature was within the normal range (39-40°C) while ruminal pH was lower than those recommended as an optimum pH of 6.5-7 for rumen fermentation when roughage was fed as major component of the diet [26]. The change in pH reported in this experiment could be due to the relatively high concentrate intake. The proportion of acetic acid and the C2:C3 ratio in control group was higher than in CH supplemented group ( $P < 0.05$ ) while propionic acid proportion was higher in CH supplemented group ( $P < 0.05$ ). This result indicates that rumen fermentation efficiency

of CH group was higher than in the control group. Condensed tannin in plants can reduce methane production in the rumen [19]. However, methane concentrations calculated in this study were not significantly lowered for CH supplementation. Condensed tannin reduced ruminal CH<sub>4</sub> due to its effect on reducing ruminal protozoa and methanogenic bacteria. However, it was not present in this study. This may be due to low intake of CT by dairy cows which range from 0.54-0.58 % of total intake.

TABLE IV. EFFECT OF CASSAVA HAY SUPPLEMENTATION ON RUMEN FERMENTATION

Items	Control	Cassava hay supplementation	SEM
Ruminal pH	6.3	6.4	0.1
Rumen temperature, °C	39.0	40.0	2.3
Total volatile fatty acid, mmol/L	104.6	102.2	4.9
	-----% of total VFA-----		
Acetate (C2)	68.0 <sup>a</sup>	65.4 <sup>b</sup>	1.6
Propionate (C3)	23.2 <sup>a</sup>	25.6 <sup>b</sup>	1.6
Butyrate (C4)	8.8	8.9	1.9
C2:C3	2.9 <sup>a</sup>	2.6 <sup>b</sup>	0.3
Methane, mol% <sup>1</sup>	25.7	22.6	1.6

<sup>1</sup> Calculated using equation of Ørskov (1992): CH<sub>4</sub> = 0.5(C2)-0.5(C3)+0.25(C4)

<sup>ab</sup> values with different superscripts differ ( $P < 0.05$ )

Chemical composition of manure-based fertilizer is shown in Table 5. Different manure management strategies did not affect organic matter and P content of the fertilizers. Manure management strategies (covered and covered with EM treated) did not reduce N and P losses during preservation when compared with non-covered group. However, crude protein and N composition in the covered manure group tended to be higher than the non-covered manure group ( $P = 0.07$ ). EM-treated, covered-manure decreased crude protein and N contents in the fertilizer when compared with the covered only group ( $P < 0.05$ ). In general, micro organisms can use N in the manure as a source for protein synthesis, thus this N in the manure is lost to the air so the CP content in this treatment group was lower than in other groups.

TABLE 5. EFFECTS OF MANURE MANAGEMENT STRATEGIES ON FERTILIZER CHEMICAL COMPOSITION

Chemical composition	Treatment <sup>1</sup>			SEM	Contrasts	
	A	B	C		A vs BC	B vs C
	----- % of DM -----					
Organic matter	38.7	41.8	42.7	3.27	ns	ns
Crude protein	7.1 <sup>ab</sup>	8.5 <sup>a</sup>	6.8 <sup>b</sup>	0.50	ns	*
Nitrogen	1.14	1.36	1.09	0.09	ns	*
Phosphorus	0.46	0.44	0.44	0.02	ns	ns

<sup>1</sup> A=fertilized with non-covered manure, B=fertilized with covered manure, C=fertilized with effective micro organisms (EM) treated with covered manure

<sup>ab</sup> values with different superscripts differ ( $P < 0.05$ )

\*  $P < 0.05$ , ns = non-significant different

Table 6 shows crop yields including CH, stylo legume hay, and cassava root from the different manure management strategies (control, covered, and EM treated-covered) that were used as fertilizer treatments. First harvest yields of total crop yield, cassava hay and legume were similar across manure managements. Cassava hay yield in the second and third periods and stylo legume yield in all periods that received treatments were not

affected by manure management strategies. However, CH production and cassava root yield in B and C groups were higher than non-covered manure group. This result indicates that cassava was more sensitive to fertilizer quality than stylo legume. Thus, by covering manure and treating cover-manure with EM will lead to increased cassava production. Stylo legume yield of EM-treated manure group was not different from the covered manure group under this study. However, cumulative yield of total crop yield and cassava hay were lower in EM-treated manure group when compared with covered manure group ( $P < 0.05$ ). It should also be noted that using EM may increase cost of crop production.

TABLE 6. EFFECTS OF MANURE MANAGEMENT STRATEGIES ON CASSAVA AND STYLO LEGUME YIELD

Items	Treatment <sup>1</sup>			SEM	Contrast	
	A	B	C		A vs BC	B vs C
Total crop yield, ton DM/ha						
1 <sup>st</sup> harvest (non-fertilized)	12.7	13.4	11.1	1.16	ns	ns
2 <sup>nd</sup>	17.0	21.6	17.5	1.44	ns	ns
3 <sup>rd</sup>	10.1	10.1	8.4	0.98	ns	ns
4 <sup>th</sup>	16.8	18.4	19.1	1.47	ns	ns
Cumulative yield	56.5	63.5	56.1	2.03	ns	*
Cassava hay, ton DM/ha						
1 <sup>st</sup> harvest (non-fertilized)	12.1	12.7	10.4	1.16	ns	ns
2 <sup>nd</sup>	4.5	5.1	4.2	0.65	ns	ns
3 <sup>rd</sup>	4.7	4.8	3.1	0.65	0.07	ns
4 <sup>th</sup>	4.0 <sup>a</sup>	5.3 <sup>ab</sup>	5.7 <sup>b</sup>	0.55	*	ns
Cumulative yield	25.3	27.9	23.4	1.48	ns	*
Stylo legume, ton DM/ha						
1 <sup>st</sup> harvest (non-fertilized)	0.53	0.68	0.66	0.10	ns	ns
2 <sup>nd</sup>	12.5	16.5	13.4	1.82	ns	ns
3 <sup>rd</sup>	5.4	5.3	5.3	0.52	ns	ns
4 <sup>th</sup>	12.8	13.1	13.4	1.70	ns	ns
Cumulative yield	31.3	35.6	32.7	1.04	ns	ns
Cassava root, ton/ha (Fresh)	19.9 <sup>a</sup>	25.3 <sup>b</sup>	27.0 <sup>b</sup>	1.48	*	Ns

<sup>1</sup> A=fertilized with non-covered manure, B=fertilized with covered manure, C=fertilized with effective microorganisms (EM) treated with covered manure

<sup>ab</sup> values with different superscripts differ ( $P < 0.05$ )

\*  $P < 0.05$ , ns = non-significant different

Dairy farmers participated in this project showed a great interest and enthusiasm in the project's activities. These farmers will transfer this technology to other farmers by an exchanging experiences and knowledge. Moreover, most of them appreciated the value of using CH as an energy and protein supplementing source for their dairy cows in order to reduce the costs of production. Issues of environment were also appreciated but of less concern to farmers than productivity. It has been well accepted that CH could be produced on farms and used by dairy farmers and should be further advanced as a feed for dairy cattle.

#### 4. CONCLUSIONS

CH supplementation tended to decrease methane production while milk yield and compositions were not affected by feeding regimes. Nitrogen and P excretions were not

decreased by CH supplementation. Manure management strategies did not reduce N and P losses during preservation. EM-treated, covered-manure decreased the N content in manure fertilizer. Based on these results, it could be recommended that CH supplementation was advantageous and manure preserving with covering should be a strategy considered to reduce methane, N, and P excretion in smallholder-dairy farms.

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# IMPROVING PRODUCTIVITY AND REDUCING METHANE PRODUCTION AND N AND PHOSPHOROUS EXCRETION IN LAMBS THROUGH NUTRITIONAL STRATEGIES.I. PRE-TREATMENT

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## Abstract

A series of trials were conducted to evaluate the effect of ammonium bicarbonate pre-treatment on nutritive value of soybean hull and to compare the performance of growing sheep offered pelleted untreated (USH) and ammoniated soybean hull (ASH) at different ratios to concentrate. In trial 1, the soybean hull was first pre-treated with ammonium bicarbonate. Three pelleted soybean hull-based diets were formulated with different ratios of concentrate mixture: (1) 60% USH and 40% concentrate (USH60), (2) 60% ASH and 40% concentrate (ASH60), and (3) 70% ASH and 30% concentrate (ASH70). Nutritive value was evaluated by *in vitro* gas test. Neutral detergent fibre (NDF) of soybean hull was reduced by 4.7% ( $P < 0.01$ ), and N (N) content increased by 70.1% ( $P < 0.01$ ) by ammonification. Compared to the USH, gas production and estimated *in vitro* organic matter digestibility (IVOMD) of ASH were significantly improved ( $P < 0.01$ ). The IVOMD was significantly higher ( $P < 0.01$ ) in diet ASH60 than that in USH60 and ASH70. Rate constant of digestion was lower in USH60 than that in the two ASH-based diets. In trial 2, twenty-seven growing Huzhou lambs (Body weight?) were randomly divided into three groups, and allocated to one of the three diets: USH60, ASH60 and ASH70. Feed intake, average body gain and feed conversion ratio were significantly higher ( $P < 0.01$ ) in ASH60 than in USH60. In trial 3, manures from trial two was gathered and manures from each pen divided into 2, one covered with polyethylene and the other without. Nitrogen content of the manures from ASH60 animals was the highest, followed by USH60, with the lowest N for ASH70, indicating that lower concentrate in the diet may reduce the N excretion in manure. After composting, N content in the covered treatments tended to be higher than that in the uncovered. The recovery of N was significantly ( $P < 0.01$ ) higher in the covered compared to the uncovered manures. Cover treatment did not have significant effect on P recovery. These results suggest that pre-treatment with ammonium bicarbonate could improve the nutritive value of soybean hull and increase the growth potential in lambs. Nitrogen loss during composting could be reduced by covering the manure with plastic.

## 1. INTRODUCTION

The major limitation to ruminant production in many Asian countries is poor nutrition. The productivity of animals is restricted by the low nitrogen (N) and high fibre content of the native forages and crop residues, which is exacerbated, by seasonal availability of these feeds, which form the basis of the diets. Chemical treatment of fibrous feedstuffs, supplementation of tropical roughages with leguminous forages and low-cost N sources, and use of agricultural by-products are promising methods to alleviate nutrient deficiencies associated with these basal diets [1, 2]. On the other hand, the N in faeces (manure) can be better utilized for crop production whereas N in urine is lost into the environment as a gas or leaches into the soil; both these phenomena cause environmental pollution. In the tropics, 80 % of the urine N is lost into the environment. The objective of this study was to improve the nutritive value of soybean hull and

enhance animal productivity through ammonia-fication and diet formulation and to decrease manure N and P (P) loss into the environment by management strategies.

## 2. MATERIAL AND METHODS

### 2.1. Ammonia treatment of soybean hull

Soybean hull (USH), processed with braker machine was purchased from Hangzhou Zhengxing Animal Industries, Zhejiang China. Ammoniated soybean hull (ASH) was prepared according to stock method [3]. One tonne of the chopped rice straw (RS) was treated with 120 kg ammonium bicarbonate (17.5% N) and 450 kg water for 30 days at ambient temperature of 15 to 20°C. The treated hull was exposed to the air for a minimum of 24 h before feeding to animals to allow free ammonia to escape.

### 2.2. Formulation of pelleted soybean hull-based diets

three soybean hull-based experimental diets were designed: 60% USH plus 40% concentrate (USH60), 60% ASH plus 40% concentrate (ASH60), and 70% ASH and 30% concentrate (ASH70). Diet ASH60 was designed to investigate the effect of ammonia-fication of soybean hull, and diet ASH70 to see the effect of replacing 10% concentrate with ASH.

To increase feed intake and minimize feed loss, all the diets were pelleted. The hulls were ground using a hammer mill (Jinshan Yongshun Industry and Commercial Co., Shanghai, China) and after thorough mixing, all the ingredients were put into a pelleting machine (Model KL-A2, Yuyao Shuanghe Feed Mill, Zhejiang). The pellets were 5 to 10 cm long with diameter of 10 mm. Two hundred grams of the pellets were sampled, ground through 40 mesh, and stored for later analysis. The pellets were analyzed for crude protein (CP), Ca and P according to the method by AOAC [4], and neutral detergent fibre (NDF) following the procedures by Van Soest et al., [5]. The ingredients and chemical composition are showed in Table I.

### 2.3. Nutritional evaluation of soybean hull-based diets

*In vitro* gas production test was used to estimate the *in vitro* digestibility. The incubations were carried out using the Reading Pressure Technique (RPT) [6]. About 750 mg of substrates were carefully weighed into 180-ml serum bottles and 90 ml of the reducing buffer medium [7] added to each bottle. The bottles were then sealed with a butyl rubber stopper and stored overnight at 4°C. Ruminant fluid was obtained from three donor sheep that were fed twice daily a mixed diet (60% grass hay and 40% concentrate) at 1.3 times maintenance. Rumen fluid was collected before the morning feeding and strained through four layers of cheesecloth and equal volumes of fluid from each sheep composited. The fluid was then held under CO<sub>2</sub> in a water-bath at 39°C until used.

Each bottle (pre-warmed to 39°C) was then injected with 10 ml prepared rumen fluid through the stopper using a needle. The displaced gas was allowed to escape before the needle was removed. The bottles were then swirled to mix the contents and placed in an incubator at 39°C. Four bottles (replicates) were used for each ration and gas pressure was recorded at 2, 4, 6, 8, 12, 24, 36 and 48 h of incubation. At the time of gas production (GP) measurement, a pressure transducer interfaced connected to a portable computer (PC) allowed the accumulated headspace gas pressure reading to be directly entered into the PC. The pressure measurements were then used to estimate the generated gas volumes [6, 7].

TABLE I. COMPOSITION AND CHEMICAL COMPOSITION OF PELLETTED USH AND ASH DIETS

	USH60 <sup>1</sup>	ASH60	ASH70
<i>Ingredients (%)</i>			
Soybean hull	60.0	0	0
Ammoniated soybean hull	0	60.0	70.0
Ground corn	19.5	19.5	14.5
Wheat bran	10.5	10.5	7.8
Soybean meal	3.9	3.9	2.9
Rapeseed meal	5.1	5.1	3.8
Lime	0.20	0.20	0.20
Minerals	0.20	0.20	0.20
Salt	0.50	0.50	0.50
Vitamin premix	0.10	0.10	0.10
<i>Chemical composition</i>			
ME (MJ/kg)	7.2	7.7	7.2
CP (% DM)	10.1	11.7	10.8
Ca (% DM)	0.53	0.53	0.59
P (% DM)	0.30	0.30	0.26

<sup>1</sup> USH60, ASH60 and ASH70 indicate that untreated and ammoniated soybean hull account for 60, 60 and 70 % of the diets.

In each incubation run, three blanks were included to correct for GP from endogenous substrates. To describe the dynamics of GP over time, the following equation [8] was chosen:  $GP = a + b(1 - e^{-ct})$ , where GP = cumulative gas production (ml),  $(a + b)$  = potential gas production (ml/g),  $c$  = rate of gas production (ml/h), and  $a$ ,  $b$  and  $c$  are constants. *In vitro* organic matter digestibility (IVOMD) of the three diets was estimated from the GP at 24 hr of incubation according to the equations by Menke and Steingass [9].

#### 2.4. Feeding trial

The feeding trial was carried out at the Jiahai sheep farm in Haining, Zhejiang, China. Twenty-seven growing male Huzhou lambs at the age of 2.5 months with initial live weight of 14 to 16 kg, were divided into three equal groups of 9 lambs each according to age, body weight and genotype. Lambs in each group were kept in three pens (three lambs per pen) with crack floor, and randomly assigned to the treatments (USH60, ASH60 and ASH70). All animals had free access to water and were fed twice daily at 0830 and 1630.

The trial lasted for 11 weeks with the first three weeks for adaptation and the subsequent 8 weeks for measurements. All animals were treated with anthelmintics during the adaptation period. In order to determine feed intake, the amounts of feeds offered and refused by the animals were measured for consecutive three days at intervals of one week. The lambs were weighed on arrival, before the morning feeding and at the beginning and end of the feeding trial and at 2-week intervals during the trial. Each weighing was conducted for two consecutive days.

A polyethylene film was lined under the pens to collect manure for a trial of composting. Twice during the measurement period, the manure was collected and weighed. The sum of the two collections was considered the total manure excreted. The

manure samples were analyzed for dry matter (DM), N and P according to the method by AOAC [4].

## 2.5. Manure composting

According to the grouping during the feeding trial, nine manure collections were obtained from the first collection. Thirty kilograms of wet manures from each group was divided into two parts (15 kg each) and composted on the polyethylene films with or without polyethylene film cover. The composting lasted for 30 days and the samples were then taken for later analysis.

The manures before and after composting were analyzed for DM, N and P according to the method by AOAC [4]. Recovery of DM, N and P was calculated as below:

$$\text{DM recovery (\%)} = (W2 \times \text{DM2}) / (W1 \times \text{DM1}) \times 100$$

$$\text{N recovery (\%)} = (W2 \times \text{DM2} \times \text{N2}) / (W1 \times \text{DM1} \times \text{N1}) \times 100$$

$$\text{P recovery (\%)} = (W2 \times \text{DM2} \times \text{P2}) / (W1 \times \text{DM1} \times \text{P1}) \times 100$$

Where, W1 and W2, DM1 and DM2, N1 and N2, and P1 and P2 are the wet weight of manure, content of DM, N and P in manure before and after composting, respectively.

## 2.6. Statistical analysis

The effects of chemical treatments on chemical composition and GP, and of different diets on growth performance were analyzed by the general linear model (GLM) procedure of SAS [10]. 2-way analysis of variance was applied to analyse the effect of composting methods on recovery of nutrients [10]. The difference of means for the treatments was tested by using Duncan's new multiple range test.

# 3. RESULTS

## 3.1. Chemical composition

Ammonia treatment increased the N contents of soybean hull by 68% ( $P < 0.01$ ), and reduced NDF content by 5% ( $P < 0.01$ ) compared to the USH (Table II). Inclusion of ASH to replace USH in the diets resulted in higher CP content but lower NDF content in ASH60 ( $P < 0.01$ ), while increasing level of ASH increased contents of NDF and Ca but decreased P content ( $P < 0.01$ ).

## 3.2. Nutritive value estimated from in vitro gas production

Cumulative gas production for USH, ASH and the three diets are presented in Figure 1. The ASH had similar GP pattern to the USH, but the gas values at all incubation times were higher in ASH than in USH, especially at later times.

TABLE II. CHEMICAL COMPOSITION OF DIFFERENT SOYBEAN HULLS AND DIETS

Feeds <sup>1</sup>	DM (%)	CP (%DM)	OM (%DM)	NDF (%DM)	Ca (%DM)	P (%DM)
<i>Soybean hull</i>						
USH	91.7	5.2 <sup>b</sup>	ND	78.9 <sup>a</sup>	0.69	0.13
ASH	90.7	8.8 <sup>a</sup>	ND	75.2 <sup>a</sup>	0.68	0.12
SEM	0.29	0.04		0.43	0.011	0.002
<i>Diets</i>						
USH60	89.4	11.1 <sup>b</sup>	93.1	51.9 <sup>b</sup>	0.50 <sup>b</sup>	0.34 <sup>a</sup>
ASH60	88.2	12.7 <sup>a</sup>	92.6	50.6 <sup>c</sup>	0.53 <sup>b</sup>	0.35 <sup>a</sup>
ASH70	89.4	11.2 <sup>b</sup>	93.0	60.4 <sup>a</sup>	0.60 <sup>a</sup>	0.29 <sup>b</sup>
SEM	-	0.06	0.12	0.10	0.006	0.003

<sup>a, b, c</sup> Means with different superscripts within the same row differ at  $P < 0.01$ .

<sup>1</sup>USH and ASH represent untreated and ammoniated soybean hull; USH60, ASH60 and ASH70 indicate that untreated and ammoniated soybean hull account for 60, 60 and 70 % of the diets.

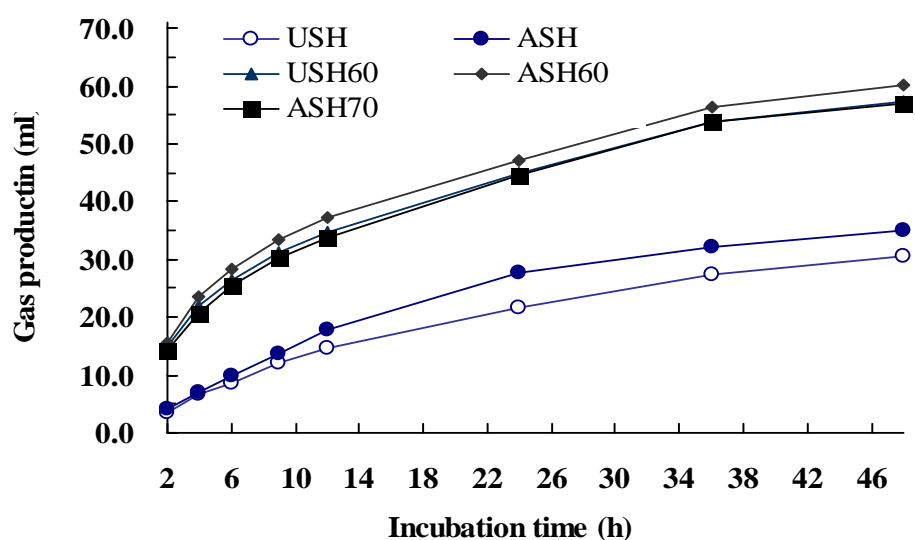


FIG 1. Gas production curve of different soybean hull diets. USH and ASH represent untreated and ammoniated soybean hull; USH60, ASH60 and ASH70 represent untreated and ammoniated soybean hull at 60, 60 and 70 % of the diets, respectively.

The ASH60 treatment had higher GP than all other diets throughout. However, there were no differences between USH60 and ASH70 suggesting that replacement of 10% concentrate mixture with ASH did not have adverse effect on the digestibility of hull diets. *In vitro* GP parameters and IVOMD are presented in Table III. The GP values at 24 and 28h were significantly higher in ASH than in USH ( $P < 0.01$ ). Potential GP ( $a+b$ ) showed higher tendency and the rate constant of GP ( $c$ ) was higher ( $P < 0.01$ ) in ASH, compared with the USH. The IVOMD was significantly higher (19%) in ASH than in USH ( $P < 0.01$ ), indicating that ammonia treatment can greatly improve the nutritive value of soybean hull. Diet ASH60 had significantly higher value of GP ( $P < 0.01$ ), potential GP ( $P < 0.01$ ), rate of GP ( $P < 0.05$ ), and IVOMD ( $P < 0.05$ ) compared to the other two diets. Replacement of 10% concentrate mixture with ASH (Diet ASH70) did not have adverse effect on all the parameters in USH60.

TABLE III. GAS PRODUCTION PARAMETER AND ORGANIC MATTER DIGESTIBILITY (OMD) FOR UNTREATED AND AMMONIATED SOUBEAN HULL AND DIETS

Feeds <sup>1</sup>	Gas production (ml)		Gas parameters		OMD (%)
	24 h	48 h	Potential (ml)	Rate (ml/h)	
<i>Soybean hull</i>					
USH	21.5 <sup>b</sup>	30.5 <sup>b</sup>	35.2	0.042 <sup>b</sup>	41.2 <sup>b</sup>
ASH	27.8 <sup>a</sup>	35.1 <sup>a</sup>	38.5	0.053 <sup>a</sup>	49.0 <sup>a</sup>
SEM	0.55	1.09	1.17	0.001	
<i>Diets</i>					
USH60	44.7 <sup>B</sup>	57.1 <sup>B</sup>	56.5 <sup>AB</sup>	0.077 <sup>b</sup>	61.8 <sup>b</sup>
ASH60	47.1 <sup>A</sup>	60.2 <sup>A</sup>	58.1 <sup>A</sup>	0.088 <sup>a</sup>	65.2 <sup>a</sup>
ASH70	45.0 <sup>B</sup>	57.2 <sup>B</sup>	55.8 <sup>B</sup>	0.083 <sup>ab</sup>	62.2 <sup>b</sup>
SEM	0.38	0.32	0.31	0.002	

<sup>a, b</sup> Means with different superscripts within the same row differ at  $P < 0.01$ ; <sup>a, b</sup> Means with different superscripts within the same row differ at  $P < 0.05$ .

<sup>1</sup> USH and ASH represent untreated and ammoniated soybean hull; USH60, ASH60 and ASH70 indicate that untreated and ammoniated soybean hull account for 60, 60 and 70 % of the diets.

### 3.3. Production performance of animals

Cumulative body weights of growing Huzhou lambs offer different diets are shown in Figure 2. There was no difference in body weight between the three groups during the first two weeks, but after that, the animals offered ASH60 had the highest weight, followed by USH60 and lastly ASH70.

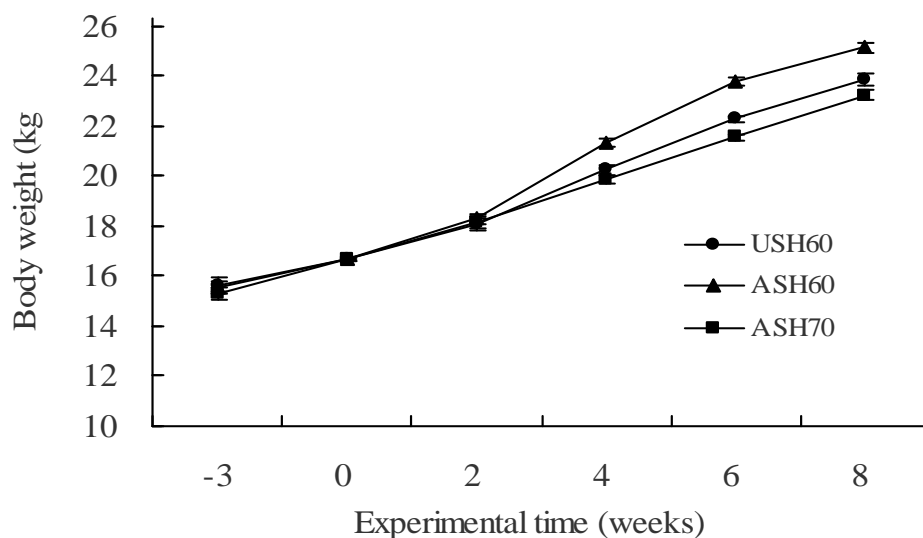


FIG 2. Cumulative body weight of sheep fed on different soybean hull diets. USH60, ASH60 and ASH70 indicate that untreated and ammoniated soybean hull account for 60, 60 and 70 % of the diets.

Table IV summarizes the feed intake and daily body weight gain of lambs. Inclusion of ASH to replace USH resulted in numeric increase in feed intake. Feed intake was not altered when ammoniated hulls were replaced by USH with lowered concentrate mixture (ASH70 vs. USH60). Daily weight gain was the highest in lambs offered ASH60 ( $P < 0.01$ ), followed by USH60 ( $P < 0.05$ ), and lowest for ASH70. Feed conversion ratio

was significantly lower in ASH60 compared to USH60 and ASH70 ( $P < 0.01$ ), while the concentrate consumption per kg of gain was the lowest in ASH70.

TABLE IV. FEED INTAKE AND AVERAGE DAILY GAIN OF HUZHOU SHEEP OFFERED DIFFERENT SOYBEAN HULL-BASED DIETS

	USH60	ASH60	ASH70	SEM
Initial body weight (kg)	16.7	16.7	16.7	0.07
Feed intake(g/d)	954	1013	929	37.2
Daily weigh gain (g/d)	128 <sup>ab</sup>	152 <sup>a</sup>	117 <sup>b</sup>	2.9
Feed conversion ratio (kg/kg)				
Total intake / gain	7.45 <sup>a</sup>	6.67 <sup>b</sup>	7.93 <sup>a</sup>	0.223
Concentrate / gain	2.98	2.67	2.38	

<sup>a, b</sup> Means with different superscripts within the same row differ at  $P < 0.01$ ; <sup>a, b</sup> Means with different superscripts within the same row differ at  $P < 0.05$ .

### 3.4. Economical evaluation

The results of the economic evaluation are shown in Table 5. Inclusion of ASH to replace USH in the diet (ASH60) resulted in the highest gross profit ( $P < 0.01$ ). Replacement of 10% concentrate mixture with ASH (ASH70) did not affect the gross profit compared to USH60.

TABLE 5. ECONOMIC ANALYSIS OF REARING LAMBS ON DIFFERENT DIETS

Diets	Weight gain (g/day)	Feed cost (Yuan/kg)	Profit (Yuan/head/day) <sup>1</sup>		
			Feed cost	Gain income	Gross profit
USH60	128	1.44	1.37 <sup>a</sup>	1.74 <sup>ab</sup>	0.37 <sup>b</sup>
ASH60	152	1.48	1.50 <sup>a</sup>	2.06 <sup>a</sup>	0.56 <sup>a</sup>
ASH70	117	1.30	1.21 <sup>b</sup>	1.59 <sup>b</sup>	0.38 <sup>b</sup>
SEM	2.9	-	0.054	0.038	0.043

<sup>a, b</sup> Means with different superscripts within the same column differ at  $P < 0.05$ ;

<sup>1</sup> The price of lambs was equal to 13.60 Chinese Yuan per kg (1USD=7.1 Chinese Yuan)

### 3.5. Manure weights and recovery of N and P in manures during composting

Amount of manure collected during the feeding trials is shown in Table 6. The animals offered ASH60 and ASH70 excreted more manure than those on USH60. Manure produced per kg of weight gain was lowest for ASH60, followed by the USH60 and highest for ASH70.



TABLE 6. MANURE WEIGHTS OF HUZHOU SHEEP OFFERED DIFFERENT SOYBEAN HULL-BASED DIETS

	USH60	ASH60	ASH70
<i>Manure excreted (kg)</i>			
Wet weight	202.3	216.9	195.2
Dry weight	53.4	58.0	59.3
<i>Manure per kg of gain (kg)</i>	7.45	6.82	9.03

Results of N and P recovery of manure under different composting methods are shown in Table 7. The N and P content in the ASH60 was the highest, followed by the USH60, and the lowest was ASH70, indicating that lowered concentrate in the diet may reduce N and P excretion in manure. After composting, N content in the covered treatments tended to be higher than that in the uncovered treatments. The recovery of N was significantly ( $P < 0.01$ ) higher in the covered than in the uncovered manures. There was no difference in P recovery.

TABLE 7. RECOVERY OF N AND P OF MANURE UNDER DIFFERENT COMPOSTING REGIMES

Items <sup>1</sup>	Uncovered			Covered			SEM	P value <sup>2</sup>		
	USH60	ASH60	ASH70	USH60	ASH60	ASH70		C	D	C*D
<i>Before composting</i>										
W1 (kg)	15.1	15.1	15.0	15.1	15.1	15.0				
DM1 (%)	26.4	26.8	30.4	26.4	26.8	30.4	0.75			
N1 (%DM)	2.0	2.2	1.9	2.0	2.2	1.9	0.17			
P1 (%DM)	0.49	0.50	0.42	0.49	0.50	0.42	0.007			
<i>After composting</i>										
W2 (kg)	20.8	21.4	21.9	21.2	21.3	22.4				
DM2 (%)	16.9	17.1	17.1	16.6	17.0	17.6	0.50			
N2 (%DM)	1.4	1.7	1.4	1.7	1.9	1.7	0.30			
P2 (%DM)	0.51	0.48	0.44	0.51	0.51	0.45	0.014			
<i>Recovery (%)</i>										
Dry matter	88.1 <sup>ab</sup>	91.2 <sup>a</sup>	82.5 <sup>c</sup>	88.1 <sup>ab</sup>	90.0 <sup>a</sup>	86.0 <sup>b</sup>	1.26	0.47	0.01	0.19
Nitrogen	64.7 <sup>b</sup>	67.3 <sup>b</sup>	61.6 <sup>b</sup>	74.1 <sup>a</sup>	75.0 <sup>a</sup>	76.0 <sup>a</sup>	1.75	0.00	0.39	0.18
Phosphorus	90.0	88.6	84.8	88.5	88.5	89.4	1.00	0.26	0.13	0.03

<sup>a, b</sup> Means with different superscripts within the same row differ at  $P < 0.01$ .

<sup>1</sup> W1 and W2, DM1 and DM2, N1 and N2, and P1 and P2 are the wet weight of manure, content of DM, N and P in manure before and after composting

<sup>2</sup> C = Cover effect; D = Diet effect; and C × D = Interaction effect between cover and diet.

## 4. DISCUSSION

### 4.1. Effect of treatment with ammonium bicarbonate on soybean hull

It is well known that treatment with ammonia sources can alter the cell wall structure and reduce the structural carbohydrates of crop residues such as cereal straws, stovers and hulls, and increase the N content of the treated materials [11, 12, 13, 14, 15,

16]. Anhydrous ammonia and urea are two sources of ammonia that are widely used. However, the use of anhydrous ammonia is unlikely to be widely accepted in China because of its high price and difficulties in transportation and handling. Urea is a chemical fertilizer in short supply and extensively used in agricultural production, which greatly influences market prices. Therefore, Ammonium bicarbonate, which is abundant and lower in price has been more widely accepted, especially in southern provinces [17, 18, 19].

Treatment with ammonium bicarbonate significantly increased N and reduced NDF content of soybean hull (Table II). The nutritional value of soybean hull was greatly improved, as reflected in the potential extent of digestion and *in vitro* OMD (Table III). Rate of digestion of soybean hull was also improved significantly ( $P < 0.01$ ). These results indicate that ammonium bicarbonate was effective in upgrading soybean hull.

When the ASH was included into the diet to replace the USH, the chemical compositions of the diets were similar to the hull (Table II). Diet ASH60 had higher potential extent of digestion, *in vitro* OMD and rate of digestion, compared to other two diets, but USH60 and ASH70 were not different from each other suggesting that the inclusion of ammoniated hull in the diet may reduce the amount of concentrate in the mixture without adverse effects on the nutritive value of diets.

#### **4.2. Effect of feeding the pelleted soybean hull diets**

It has been demonstrated that pelleting diets may increase intake by 89% and improve feed efficiency by 28% [20]. The intake of sodium hydroxide treated straw diets in lambs was also increased by the pelleting treatment [21]. Therefore, all diets in the current study were pelleted to increase feed intake and decrease feed loss.

three soybean hull based diets were designed in this study. The diet with 60% USH and 40% concentrate (USH60) was used as control. The diet containing 60% ASH (ASH60) was designed to investigate the effects of ammoniaification whereas the diet with 70% ASH was to study the effect of replacing 10% concentrate with ASH. Compared to USH60, DM intake tended to increase for ASH70, but the digestibility was significantly increased (Table III). Replacing USH with ASH resulted in a numeric increase in intake. However, feed intake was not altered when ammoniated hull was used to replace USH with lowered concentrate (ASH70 vs. USH60). Daily weight gain was the highest in lambs offered ASH60 ( $P < 0.01$ ), followed by USH60 ( $P < 0.05$ ), and lowest for ASH70. However, the concentrate consumption per kg of gain was the lowest with ASH70.

Because the price of lambs was relatively higher at that time, the highest gross profit was obtained for the animals fed ASH60, which had the highest body weight gain. Lower use of concentrate with ASH did not increase the gross profit compared to USH60, but it saved the use of concentrate mixture, which may be used for other animals. The cost per kg of feed was the lowest for ASH70.

Which diet formulation should be chosen will depend on such factors as available feed resources, prices of feeds and price of the product be it meat or milk. When the concentrate mixture is not limited and the price of animals is relatively high, farmers may choose the formulation like ASH60. However, the formulation with lower concentrate inclusion such as ASH70 may be more suitable for the situations in the most of developing countries where the grains are usually limited but the unconventional feed resources are of abundant supply.

### 4.3. Effect of covering on loss of N and P during composting

Animal manures are usually composted before used as fertilizers so that the infectious micro organisms could be killed. Normally farmers compost the manure on their farms without any treatments. During these processes, Nour compounds will be degraded to produce ammonia, which will be vaporized into the environment (air) whereas the degradation of nutrients in the manures under anaerobic conditions may be inhibited [22, 23, 24]. In the current study, the use of polyethylene films to cover the manure during the composting significantly increased the recovery of N (Table 7), indicating that a simple use of coverage on the top of the manure can reduce the loss of N which is beneficial to the plants.

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# IMPROVING PRODUCTIVITY AND REDUCING METHANE PRODUCTION AND N AND PHOSPHOROUS EXCRETION IN LAMBS THROUGH NUTRITIONAL STRATEGIES. II. METHANE PRODUCTION

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## Abstract

Three trials were conducted to investigate the effect of tea saponins (TS) on production methane production and productive performance in growing goats. In trial 1, TS was added at 0, 0.2, 0.4 and 0.8 mg/ml to study the effects on the ruminal fermentation *in vitro*. Methane production was significantly decreased when 0.4 and 0.8 mg/ml of TS was added. The TS had little effect on ruminal pH and total volatile fatty acids. However, the fermentation patterns were changed, with lower proportion of acetate and higher proportions of propionate when TS was added. Ammonia-N concentration and protozoal counts were significantly lower with TS addition, while microbial protein yield increased suggesting that the TS could modify ruminal fermentation. Twenty-seven growing Boer goats were used in trial two to evaluate the effects of TS addition on growth performance. The animals received the same basal diet (300g concentrate mixture, 200g alfalfa and corn stover silage *ad libitum*) and TS was added at 0, 3 and 6 g per day. Dry matter intake, average daily gain and feed efficiency in the 3 g TS group were higher than in the other two groups. Trial three was conducted to investigate the effects of TS (5 g/kg DM), TS plus disodium fumarate (DF) (20 g/kg DM) and coconut oil (7% on DM basis) on methane production in sheep using a simple box chamber. Eight Huzhou sheep were assigned to 4 treatments in a 4 × 4 Latin square design. Addition of TS and TSDF decreased ( $P < 0.0001$ ) methane production by 8.5 and 9.6%, respectively. Addition of TSDF increased ( $P < 0.01$ ) the proportion of propionate and decreased ( $P < 0.01$ ) the proportion of acetate. These results suggest that addition of TS and TSDF, though not as effectively as coconut oil, can inhibit methane production, which is beneficial for economy and environment.

## 1. INTRODUCTION

Recent studies in respiration chambers have confirmed that methane production from ruminants fed on fibrous diets are higher than methane outputs from better quality temperate forages. Livestock produce approximately 80 to 115 million tons of methane per year, equivalent to 15 to 20% of total anthropogenic methane [1]. Methane produced during ruminal fermentation represents a loss of 8 to 12% of gross energy intake [2]. Therefore, inhibition of methane production can represent an improvement in feed efficiency and short-term economic and long-term environmental benefits.

Attempts to reduce methanogenesis by the addition of chemicals, such as halogen compounds and ionophores, have long been made. Ionophores, such as monensin and lasalocid, have especially been put to practical use. However, these ionophores may depress fibre digestion [3]. Because one of the greatest merits of ruminants is the ability to utilize fibre, methane production should be reduced without depressing fibre digestion.

Fumarate has been shown to be an effective suppressant of methanogenesis [4], and saponins extracted from tea seed had a beneficial effect *in vitro* [5]. It hypothesized that the methane suppressing effects of tea saponins would lead to an accumulation of hydrogen, which would be removed by addition of fumarate, since hydrogen can act as a

sink when fumarate is reduced to succinate [6]. On the other hand, the N (N) in faeces (manure) can be better utilized for crop production whereas N in urine is lost into the environment as a gas or leaches into the soil; both the phenomena causing environmental pollution. In the tropics, 80 % of the urine N is lost into the environment. Therefore, the objective of this study was to reduce methane production from ruminants using saponins extracted from tea seed.

## 2. MATERIAL AND METHODS

### 2.1. *In vitro* rumen fermentation

#### 2.1.1 *Materials.*

The substrate used in the incubation study was a mixture of grass meal and corn meal (50:50 on a w/w basis). Tea saponin (TS) used in this study was purchased from Zhejiang Orient Tea Development Co., Ltd. an affiliated of the Institute of Tea Research, Chinese Academy of Agricultural Sciences. It was in powder form with light-yellow colour, and contained 60% triterpenoid saponin.

#### 2.1.2 *Experimental design.*

The TS was added at 0, 0.2, 0.4 and 0.8 mg/ml in the *in vitro* rumen fluid. The incubations were carried out using the RPT (Mauricio et al., 1999). Four replicate bottles were used for each level of TS and incubated for 3, 6, 9, 12 and 24 h. Gas pressure was recorded and a sample of the gas was drawn out using a needle through the stopper for methane concentration determination using a GLC. At 24 h incubation, the incubation was stopped and the ruminal fluid sampled to determine pH, ammonia-N, volatile fatty acids (VFA), protozoa counts, and microbial protein (MCP) yield. The details of the *in vitro* gas production test were as described as above.

#### 2.1.3 *Measurement of fermentation parameters.*

The pH of rumen liquor was determined immediately after removal using a pH meter (Model PB-20, Sartorius). For determination of ammonia-N and VFA concentrations, 0.25 ml of 25 % ortho-phosphoric acid was added to 1 ml of inoculum sample, the mixture was mixed uniformly and centrifuged at 10000 rpm for 10 min. The supernatant was then transferred to another test tube, capped and stored in a freezer at -20°C until analyzed. Ammonia-N concentration was determined using a spectrometer (Model 721) using colorimetry with NH<sub>4</sub>Cl solution as a standard [7]. VFA was analyzed by GLC (GC-2100, Shimadzu) equipped with a Flame Ionization detector. The column (HP-INNOWAX, 19091N-133) was 30m × 0.25mm × 0.25µm in size. two microlitres of rumen fluid samples were injected using a syringe, and the temperature of the injector/detector and the column raised to 260°C and 220°C, respectively using N as a carrier. Methane concentration was analyzed using the GLC with the same column at a temperature of the injector/detector and the column of 130°C and 80°C, respectively.

The protozoa were counted by the method of [8]. Half ml of sample was mixed with 0.5 ml methylgreen-formaldehyde-saline solution and held for 5 minutes, then pipetted into a refitted counting haemocytometer of 0.3 mm and the protozoa counted under microscope. Concentrations of the MCP was determined based on purines using the method of [9] and modified by [10], and estimated from the ratio of purines to N of isolated bacteria with yeast RNA as standard.

## 2.2. Feeding trial 1

The trial was conducted in a randomized complete block design. Twenty-seven growing Boer goats (18 male and 9 female), with an initial weight of  $17.5 \pm 2.9$  kg, were divided into three equal groups according to age, body weight and gender and each of the groups assigned to one of three treatments. Goats in each group were kept in three pens (three goats each). All animals were offered the same basal diets (300g concentrate mixture, 200g alfalfa and corn stover silage *ad libitum*) with TS added at 0 (TS0), 3 (TS3) or 6 g (TS6) per day. The amount of TS to add was calculated from the *in vitro* study above. Chemical composition (DM, CP, Ca and P) of the treatments were determined according to [11], and NDF analyzed as outlined by [12]. The ingredient and chemical composition of the concentrate is shown in Table I.

TABLE I. INGREDIENTS AND CHEMICAL COMPOSITION OF THE EXPERIMENTAL FEEDS

Composition	Concentrate mixture <sup>1,2</sup>	Alfalfa	Corn stover silage
Dry matter (g/kg)	867	889	221
Crude protein (g/kg DM)	146	169	90
NDF (g/kg DM)	204	495	547
Ca (g/kg DM)	4.9	9.2	2.1
P (g/kg DM)	4.2	1.9	0.9

<sup>1</sup> Ingredients (%): corn grain 59, wheat bran 14, soybean meal 12, rapeseed meal 5, wheat midding 7.5, and minerals-vitamins mix 2.5

<sup>2</sup> Composition of minerals-vitamins mix (per kg): Zn 100 mg, Fe 90 mg, Mn 20 mg, Cu 10 mg, I 0.30 mg, Se 0.20 mg, vitamin A 5 000 IU, vitamin D<sub>3</sub> 700 IU, vitamin E 30 IU, Vitamin B<sub>12</sub> 0.03mg, riboflavin 10 mg, D-pantothenic acid 15 mg, niacin 25 mg.

The goats were fed twice daily at 07:00 and 19:00 and had free access to drinking water. The experiment lasted for 60 days with the first 15 days for adaptation and the subsequent 45 days for measurements. The goats were weighed for two consecutive days before the morning feeding at the beginning and end of the trial, and at intervals of 15 days during the trial. The amount of feeds offered and refused by the animals was recorded daily, and feed intake was measured at intervals of 5 days.

### 2.1.3. Statistical analysis

The general linear models (GLM) procedure of [13] was used to analyze the data. The data from the *in vitro* experiment were analyzed as single factorial arrangement with multiple levels in the model for fermentation parameters. The data in the feeding trial was analyzed as a randomized complete block design using GLM procedures to evaluate the effect of tea saponin on the growth performance and serum biochemical parameters. Comparisons among dietary treatments were made using the least significant difference procedure when the F-test for treatment was significant ( $P < 0.05$ ).



## 2.3 Feeding trial 2

### 2.3.1. Animals, feeds and experimental design

Eight Huzhou sheep were randomly paired (4 fistulated and 4 none fistulated). The sheep were fed a based diet of 600 g lucerne hay plus 400 g concentrate mixture (as is basis) per day. The concentrate contained (%): corn 55.8, soyabean meal 15.8, wheat 15.8, rapeseed meal 12.6). Four treatments were designed with different additives: (1) no additive (control), (2) TS (5 g/d), (3) TSDF (TS, 5 g/d plus DF, 20 g/d) and (4) coconut oil (7 % of the DM) as positive control. The basal diet was fed in control, TS and TSDF, but the diet with coconut oil was adjusted to obtain the same levels of metabolisable energy and crude protein as the other three treatments, because of the high energy content in the coconut oil. In treatment 4, corn was reduced to 12.2% of the whole diet. The TS and coconut oil was mixed with the diets before feeding. Chemical composition of the two diets is shown in Table II. The diets were given in equal portions twice a day at 08:30 and 16:30. The sheep had free access to water.

TABLE II. CHEMICAL COMPOSITION (DM BASIS) OF THE DIET USED IN EXPERIMENT

	Diet	
	1 <sup>1</sup>	2 <sup>2</sup>
<i>Chemical composition, g/kg DM</i>		
OM	944	942
CP (6.25 × N)	13.8	13.8
NDF	436	432
ADF	225	225
Ca	8.8	8.8
P	6.8	6.8
DE (MJ/kg DM)	8.03	8.03

<sup>1</sup> the diet was used for control, TS and TSDF; <sup>2</sup> the diet was used for coconut oil.

Two simple open-circuit respiratory chambers were used to measure methane production *in vivo*. The cuboid chambers (2.4 × 1.6 × 1.5 m) are constructed using concrete with an arched roof covered with plastic film. Through a pump set to the outlet, outside air was supplied to the chamber from one end and chamber air was removed from another. The sketch of the system is outlined in Figure 1. Four pairs of sheep were randomly assigned to the 4 treatments in a double 4 × 4 Latin square design and each period lasted for 21 days. Because only two chambers were available, only two pairs of animals were used at the same time. Before the morning feeding on d 15, the first two pairs of sheep were moved to one of the chambers for measurements of methane. The first day within the chambers was considered as adaptation period, allowing the sheep to adapt before measurements were recorded for two consecutive days starting at 08:00 in the morning. In the morning of d 18, another two pairs of sheep was used for measurements. When not used for methane measurements in chambers, animals were kept in individual pens.

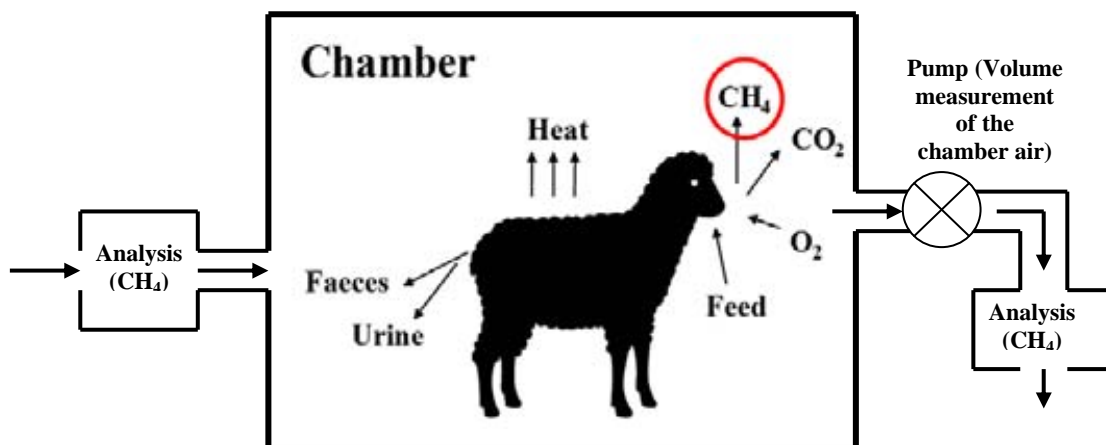


FIG. 1. Simple open-circuit respiratory chamber

### 2.3.2. Sampling procedures and measurements

During the two consecutive days when the sheep were housed in chambers, air sample was taken per hour from the exhaust pipe of each chamber with an airproof syringe, the volume of the air flowed through the chamber was recorded, and then the air sample was analysed for methane concentration by gas chromatograph (GC-2100, Shimadzu) equipped with a Flame Ionization detector (FID). After the sheep were removed from the chambers, rumen fluid samples were taken from the rumen cannulated sheep before the morning feeding, using a tube via the fistula. Immediately after collection, rumen fluid samples were strained through four layers of compress gauze. The pH of rumen liquor was determined immediately after removal using a pH meter (Model PB-20, Sartorius). VFAs were analysed using gas chromatography (GC-2100, Shimadzu). Samples were injected into a 2m × 3mm glass column packed with Porapak Q (80 mesh). Nitrogen was used as a carrier. The details were described elsewhere [14].

### 2.3.3. Statistical analysis

The results were analysed according to Latin square design for the effect of diet (n = 4), period (n = 4) and animal (n = 4) on methane production [13]. Multiple comparisons among means were carried out by the Duncan's multiple range test.

## 3. RESULTS

### 3.1. Effects of tea saponins on in vitro ruminal fermentation and growth performance in growing Boer goat

#### 3.2.1. Rumen fermentation characteristics

Addition of TS at higher level (0.8 mg/ml) decreased the 24h in vitro gas production (Table III). The OMD decreased when 0.8 ml/ml of TS was added, without significant difference between the addine levels of 0, 0.2 and 0.4 ( $P > 0.05$ ). Addition of the TS significantly decreased the methane production at 12 and 24h compared to the control (Figure 2). After 24h incubation, inclusion of 0.2, 0.4 and 0.8 mg/ml TS reduced methane production by 8.5, 15.0 and 22.7%, respectively (Table III). There was a dose-dependant manner for the TS in suppressing the methane production. These results

suggest that TS may be a good methane-inhibitor when the inclusion level is above 0.4 mg/ml in in vitro rumen fermentation.

The addition of TS reduced the final pH, but all values were in the normal range. Inclusion of TS inhibited the protozoal growth in rumen fluid. After 24h incubation, the protozoa counts were reduced by 19, 22 and 45% for 0.2, 0.4 and 0.8 mg/ml TS, respectively, from  $0.64 \times 10^5$ /ml in the control. This reducing protozoa was accompanied with the decreased ammonia-N concentration and increased MCP concentration. After 24h incubation, the MCP concentration was 32 and 39% higher than the control (0.85 mg/ml) with addition of 0.4 and 0.8 mg/ml TS, respectively. Total concentrations of VFA were not significantly influenced by the TS, but the patterns of rumen fermentation were changed, with decreased acetate proportions and increased propionate proportions.

TABLE III. EFFECTS OF TEA SAPONINS ON *IN VITRO* GAS PRODUCTION, ESTIMATED ORGANIC MATTER DIGESTIBILITY (OMD), METHANE PRODUCTION AND RUMEN FLUID CHARACTERISTICS IN THE INCUBATED RUMEN BUFFER

Items	Adding levels of tea saponin (mg/ml)				SEM
	0	0.2	0.4	0.8	
Gas production (ml)	110 <sup>a</sup>	109 <sup>a</sup>	104 <sup>ab</sup>	98 <sup>b</sup>	1.5
OMD (%)	61.4 <sup>a</sup>	60.8 <sup>a</sup>	59.3 <sup>b</sup>	57.2 <sup>c</sup>	0.50
Methane (mmol)	0.76 <sup>a</sup>	0.69 <sup>ab</sup>	0.64 <sup>bc</sup>	0.58 <sup>c</sup>	0.021
Methane (mmol/g OMD)	1.63 <sup>a</sup>	1.51 <sup>ab</sup>	1.44 <sup>bc</sup>	1.36 <sup>c</sup>	0.038
pH	6.91 <sup>a</sup>	6.86 <sup>ab</sup>	6.85 <sup>ab</sup>	6.73 <sup>b</sup>	0.036
Ammonia-N (mmol/l)	11.6 <sup>a</sup>	10.4 <sup>ab</sup>	8.9 <sup>ab</sup>	8.4 <sup>b</sup>	0.20
Total VFA (mmol/l)	49.5	52.9	52.0	52.8	1.48
Acetate (%) <sup>1</sup>	72 <sup>a</sup>	69 <sup>b</sup>	68 <sup>b</sup>	67 <sup>b</sup>	0.6
Propionate (%) <sup>1</sup>	18 <sup>c</sup>	21 <sup>b</sup>	22 <sup>b</sup>	25 <sup>a</sup>	0.5
Butyrate (%) <sup>1</sup>	10 <sup>a</sup>	10 <sup>a</sup>	10 <sup>a</sup>	8 <sup>b</sup>	0.2
Acetate/Propionate	3.98 <sup>a</sup>	3.33 <sup>b</sup>	3.08 <sup>c</sup>	2.72 <sup>d</sup>	0.091
Protozoa ( $\times 10^5$ /ml)	0.64 <sup>a</sup>	0.52 <sup>b</sup>	0.50 <sup>b</sup>	0.35 <sup>c</sup>	0.011
Microbial protein (mg/ml)	0.85 <sup>b</sup>	0.98 <sup>ab</sup>	1.12 <sup>a</sup>	1.18 <sup>a</sup>	0.058

<sup>1</sup>Percentage of the individual fatty acids to the total.

<sup>a,b</sup> Means with different superscripts within the same row differ at  $P < 0.01$ .

### 3.2.2. Growth performance

During the whole feeding period, dry matter intake (DMI) by animals in group TS3 was higher than that in C and TS6, though the difference showed no statistical significance for the intake during d 31-45 among the three groups (Table IV). The goats with 3g of TS had higher average daily gain and gain/feed than those on group TS0 and TS6 (Table IV) during the whole period. The effects were more pronounced in the later stage of the feeding experiment.

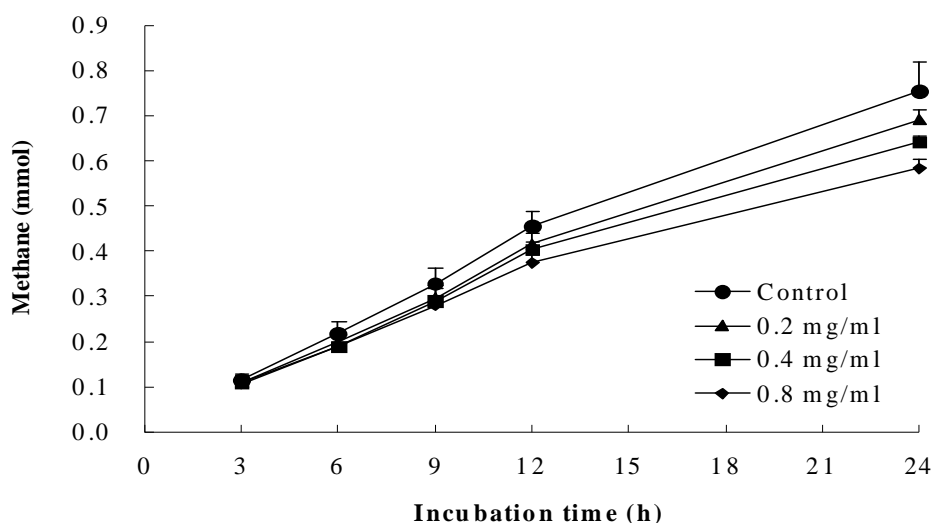


FIG. 2. Effect of tea saponin addition on methane production (Bars indicate standard error)

TABLE IV. FEED INTAKES, AVERAGE DAILY GAIN AND GAIN/FEED OF BOER GOATS FED ON A DIET SUPPLEMENTED WITH TEA SAPONIN AT 0 (TS0), 3 (TS3) OR 6G (TS6)

Items	Diets			SEM
	TS0	TS3	TS6	
Initial weight (kg)	17.0	17.7	17.7	0.31
Feed intake (g DM/day)				
0-15 d	521 <sup>b</sup>	612 <sup>a</sup>	518 <sup>b</sup>	22.5
16-30 d	537 <sup>b</sup>	639 <sup>a</sup>	536 <sup>b</sup>	18.1
31-45 d	551	617	549	18.4
0-45 d	537 <sup>b</sup>	623 <sup>a</sup>	534 <sup>b</sup>	18.6
Average daily gain (g/day)				
0-15 d	59	66	82	7.7
16-30 d	49 <sup>b</sup>	70 <sup>a</sup>	66 <sup>ab</sup>	4.6
30-45 d	93 <sup>ab</sup>	122 <sup>a</sup>	79 <sup>b</sup>	6.4
0-45 d	69 <sup>B</sup>	94 <sup>a</sup>	71 <sup>b</sup>	2.5
Gain/Feed (g/kg)				
0-15 d	111	125	134	13.7
16-30 d	91 <sup>b</sup>	109 <sup>ab</sup>	123 <sup>a</sup>	7.8
30-45 d	167 <sup>ab</sup>	196 <sup>a</sup>	143 <sup>b</sup>	10.2
0-45 d	127 <sup>b</sup>	150 <sup>a</sup>	131 <sup>b</sup>	3.8

<sup>a, b</sup> Means with different superscripts within the same row differ at  $P < 0.05$ .

### 3.2.2. Inhibition of methanogenesis by tea saponin and tea saponin plus disodium fumarate

Addition of TS, TSDF and coconut oil resulted in similar diurnal pattern of methane production as control (Figure 3). Methane production was rapidly increased to maximum 2-3 hours after the animals were fed, and then decreased slowly until the next feeding. The diets with neither TS nor TSDF have effects ( $P > 0.05$ ) on methane production in daytime, but methane production were significantly decreased ( $P < 0.001$ ) at night.

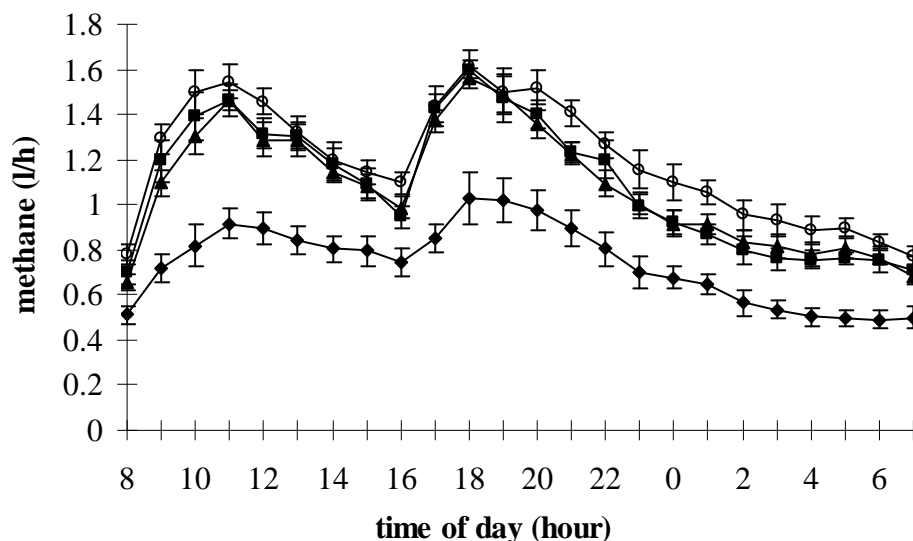


FIG. 3. Diurnal pattern of methane production from sheep in a chamber fed diets with different additives: control (-○-, no additive), tea saponin (-■-, TS 5g/kg DM), tea saponin plus disodium fumarate (-▲-, TS, 5 g/kg DM plus DF, 20 g/kg DM) and coconut oil (-◆-, 7% DM). The vertical bars indicate standard error of mean at selected times.

There was little difference in dry matter intake (not shown) through the experiment, because restricted feeding was adopted for all sheep. Methane production was reduced ( $P < 0.001$ ) by all the additives, with little difference between TS and TSDF (Table 5).

TABLE 5. RUMINAL PH AND VOLATILE FATTY ACIDS (VFAS) VARIABLES FOR SHEEP FED DIETS WITH OUT OR WITH TEA SAPONIN (TS), TS PLUS DISODIUM FUMARATE (TSDF) OR COCONUT OIL

Item	Control	TS	TSDF	Coconut oil	SEM	P-value
Methane, l/kg of DMI	28.7 <sup>a</sup>	26.2 <sup>b</sup>	25.9 <sup>b</sup>	17.7 <sup>c</sup>	0.57	<0.0001
Ruminal pH	6.82 <sup>ab</sup>	6.60 <sup>c</sup>	6.91 <sup>a</sup>	6.69 <sup>bc</sup>	0.06	0.04
Total VFAs, mmol	52.7	52.0	55.6	50.4	2.87	0.66
<i>Molar proportions, %</i>						
Acetate	69.6 <sup>ab</sup>	68.4 <sup>b</sup>	65.2 <sup>c</sup>	71.1 <sup>a</sup>	0.72	0.006
Propionate	22.0 <sup>b</sup>	22.3 <sup>b</sup>	26.3 <sup>a</sup>	21.8 <sup>b</sup>	0.55	0.003
Butyrate	8.4 <sup>a</sup>	9.3 <sup>a</sup>	8.5 <sup>a</sup>	7.2 <sup>b</sup>	0.28	0.001
Acetate: propionate	3.18 <sup>a</sup>	3.07 <sup>a</sup>	2.48 <sup>b</sup>	3.27 <sup>a</sup>	0.11	0.007

<sup>a,b,c</sup> within a row, means without a common superscript letter differ,  $P < 0.05$

Compared with the control, ruminal pH was decreased by addition of TS ( $P < 0.05$ ), but not by TSDF and coconut oil ( $P > 0.05$ ) (Table 5). Although there were no significant differences in total VFA concentrations among diets, proportions of individual VFAs differed ( $P < 0.01$ ). Addition of TS had no significant effects on VFAs. Fumarate addition resulted in a significant increase ( $P < 0.01$ ) in propionate and a significant decrease in acetate production, but did not affect butyrate production. Addition of

coconut oil showed no effects on the concentrations of acetate and propionate, but the concentrations of butyrate were significantly reduced ( $P < 0.01$ ).

#### 4. DISCUSSION

##### 4.4. Effect of tea saponin addition on growth performance and methane production in growing lambs

Addition of TS reduced the final pH in rumen *in vitro* and *in vivo* (Tables 3 and 5). Asanuma [4] concluded that if fumarate is added to ruminant feed, its sodium salt should be used because fumaric acid decreases ruminal pH. In current study, addition of DF avoided the decrease in pH *in vivo*. In effect, the sodium moiety serves as an agent to raise pH, which is similar to the effect of the addition of sodium bicarbonate. Addition of TS at higher level (0.8 mg/ml) decreased the 24h *in vitro* gas production (Table III), which is probably due to the TS-mediated inhibition of cellulolytic bacterial and fungal growth [17]. In this study, the protozoa counts were reduced significantly by TS addition. Protozoa contribute significantly to intraruminal cycling of microbial N, thus reducing protozoa in rumen could improve ruminal N utilization and increase microbial protein flow to the intestine [15, 16]. In fact, the MCP concentration was 32 and 39% higher than the control (0.85 mg/ml) with addition of 0.4 and 0.8 mg/ml TS, respectively.

The patterns of rumen fermentation *in vitro* and *in vivo* were changed due to addition of TS (Tables 3 and 5), with decreased acetate and increased propionate proportions, similar to the observations of others [10, 18]. Addition of TSDF decreased acetate concentration, but propionate was significantly increased, which may be at the expense of acetate. Newbold [6] found that both fumarate and malate are key intermediates in the succinate-propionate pathway, in which malate is dehydrated to fumarate and fumarate reduced to succinate, which is then decarboxylated to propionate.

There was a dose-dependant manner for the TS in suppressing the *in vitro* methane production, suggesting that TS may be a good methane-inhibitor when the inclusion level is above 0.4 mg/ml in *in vitro* rumen fermentation (Table III). The methane suppressing effects of saponin might be due to the inhibition of H<sub>2</sub>-producing bacteria such as cellulolytic bacteria and the elimination of protozoa [14, 17]. However, addition of TS reduced *in vivo* methane production by 8.5% (Table 5), lower than that obtained with approximately the same dose *in vitro* (Table I0). The lesser extent may be due to the flow of liquid through the rumen, which may dilute the concentrations of TS. The methane-suppressing effects of saponin were presumably a direct action against the rumen microbes involved in methane formation, including methanogens and protozoa [19].

It is presumably that the methane-suppressing effects of TS may lead to accumulations of hydrogen. Another way of diverting hydrogen away from methane formation would be to promote alternative electron-sink metabolic pathways to dispose of the reducing power [20], and fumarate has been shown to compete successfully for hydrogen [6]. However, further addition of DF did not have better inhibitory effects on methane production compared to TS addition only (Table 5). In the Rusitec, 6.25 mmol of fumarate caused a 1.2 mmol fall in methane [21]. In the current study, daily methane production were reduced about 0.45 mol by addition of TSDF in a chamber, so if DF only, almost 2 mol (more than 300 g) fumarate is needed. However, the daily dose of fumarate was 20 g/d per sheep. Consequently, the contribution of fumarate on methane inhibition is limited in this study. Fumarate would be impractical as a means of eliminating methane production *in vivo*. Newbold [6] concluded that although it might be possible to use organic acids and fumarate in particular, to decrease ruminal methane

production, the quantities required to make a major impact on daily methane production might be impractical.

Dry matter intake by animals with 3g of TS was higher than that in control and 6g of TS, though the difference showed no statistical significance for the intake during d 31-45 among the three groups (Table IV). These results are different from the results of [22], who found the decreased dry matter intake in the Yucca extracts-added diets. Other researchers reported little effects of saponin on feed intake [23, 24]. The goats with 3g of TS had higher average daily gain and gain/feed than those on 0 or 6g TS-added group (Table IV) during the whole period. The effects were more pronounced in the later stage of the feeding experiment. Improved animal performance were observed when Yucca saponins were fed to the lactating cows [24], whereas [22] suggested that saponins could not improve the animal performance when 8g/d Yucca saponins were administered to dairy cows.

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# IMPROVING GENETIC POTENTIAL AND FERTILITY OF DAIRY BUFFALO THROUGH APPROPRIATE MANAGEMENT INTERVENTIONS

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## Abstract

Delayed puberty and sub-oestrus are major reproductive disorders of economic importance in buffaloes. The genetic improvement is also slow in buffaloes mainly due to preference of the farmers for natural service with bulls of unknown genetic makeup. In the present study, a survey was conducted to identify the factors responsible for delay in puberty in dairy buffaloes. Nutritional (urea molasses multi-nutrient blocks; UMMB) and hormonal interventions (equine chorionic gonadotropin; ECG) were used to hasten the onset of puberty in dairy buffaloes. Oestrus could be induced in 40 to 100% heifers with variable conception rates (0 to 100%). 'Ovsynch' protocols of oestrus synchronization followed by fixed time artificial insemination with semen of bulls of known pedigree was applied to address the problem of sub-oestrus and poor genetic potential of the herd. Twenty-seven buffaloes that failed to exhibit oestrus up to 2 years postpartum were confirmed to be in sub-oestrus for having active corpus luteum and 1.0 ng/ml plasma progesterone concentrations were subjected to ovsynch treatment protocol. Treatment was started (day 0) irrespective of the stage of oestrus cycle on the first day of examination. 20 µg buserelin (receptal 5ml; intervet) was injected intra-muscular on day 0, followed by cloprostenol 500µg (vetmate 2ml; vet care) on day 7 and second injection of receptal (2.5 ml) on day 9. Fixed time AI was done 16 and 40 hrs thereafter. Buffaloes returning to oestrus were served naturally and pregnancy was diagnosed 90 days later. Variable signs of oestrus were observed in the buffaloes: vaginal mucus discharge in 40.7%, standing heat in 18.5%, variable degree of cervical dilatation and uterine tone in 100%. Treatment induced ovulation occurred in 88.9% buffaloes of which 51.9% conceived to the AI the overall conception rates up to three services were 66.7%. It was concluded that poor nutrition and management are major impediments for delaying onset of puberty in dairy buffaloes. Supplementary feeding of UMMB can be successfully applied to hasten onset of puberty. Ovsynch successfully synchronized ovulation in sub-oestrus buffaloes and resulted in enhanced fertility.

## 1. INTRODUCTION

Livestock forms an integral part of agriculture in India and involves participation of around 70% of its population, most of who are small/medium scale holders. Buffalo farming is an important component of Indian livestock industry and contributes more than 50 million tons of milk and 1.43 million tons of meat in addition to high valued hides, bones and draft power for agricultural operations. The buffalo holds similar importance in several other countries in Asia including Pakistan, Philippines, Sri Lanka and Nepal. However, buffalo lacked the much desired attention of the policy makers and the scientists, alike. This, along with unorganized slaughtering lead to fast degradation of buffalo genetic potential. Their number is dwindling in several countries of the world.

Poor reproduction (delayed onset of puberty, anoestrus, sub-oestrus) in buffalo is a major impediment to its efficient production. Most buffaloes (30 to 80%) do not exhibit overt oestrus especially during summer, and remain un-bred, leading to prolong infertile periods and, therefore, high economics losses. Due to rather lower success of AI in several developing countries, buffaloes there are often bred naturally with bulls of

unknown or poor genetic constitution. The present studies were designed to (i) study the factors affecting onset of puberty under field conditions in India, (ii) hasten the onset of puberty through nutritional and hormonal interventions, (iii) improve fertility in sub-oestrus buffaloes, and (iv) produce offspring with improved genetic potential.

## **2. MATERIAL AND METHODS**

### **2.1 Study on factors affecting onset of puberty in dairy buffaloes**

A survey was conducted to identify factors affecting onset of puberty in buffalo heifers. Data was collected from 1339 Murrah graded buffalo heifers on 180 rural farm spread over six agro-climatic zones of the Punjab state of Northern India. Only those heifers, which were reared by a farmer from birth were included in the study. The farmers were categorized into small, marginal and progressive, based on their herd strength of  $\leq 5$ , 5 to 30 and  $>30$  adult livestock units, respectively. Altogether, 21 factors were considered, including farmer's education, landholding and expertise in farm management, herd strength, presence of bull, housing index, feeding status of herd, season of birth, diseases/accidents at birth, and during calf-hood, deworming, suckling and/or feeding of colostrums, pre-weaning calf starters, type of calf starter and milk replacer, calves herding density, nutritional availability to the yearlings and provisions for exercise, wallowing and/or bathing of heifers. The data was analyzed statistically.

### **2.2 Effect of certain nutritional or hormonal interventions on the onset of puberty**

Ten buffalo heifers which had attained the age of three years, but not puberty, were supplemented with UMMB (Molasses 35%, Urea 10%, deoiled rice bran 17%, oiled rice barn 16%, ground nut cake 10%, cement 10%, salt 2%; prepared by cold method [1]) for 30 days during the summer season. The basal diet of these buffaloes included 25 to 30 kg green fodder (Barseem in its third cut) along with 3 to 5 kg wheat straw. Another group of 10 similar heifers were injected once with eCG (1000 IU) during the summer months. Oestral responses were recorded by the respective farmers and the heifers were mated naturally.

### **2.3 To improve fertility of the sub-oestrus buffaloes while improving the genetic potential of their offspring**

Thirty-one pleuriparous lactating buffaloes, apparently anoestrus for over 10 to 24 months after parturition were investigated in the months of December – February i.e. the favourable period for buffalo breeding in North India. These buffaloes belonged to organized dairy farms, were fed reasonably well and the oestrus detected using teaser bulls and visual observations twice daily. Each animal was fed 20 to 30 kg of seasonal green fodder and 2 to 3 kg of concentrates daily and were yielding 2 to 8 kg milk. Maximum milk yield had been up to 20 kg per day). Their body condition score varied from 2.00 to 4.25 (Basis 1.0 to 5.0). The animals were subjected to per-rectal palpation of genitalia and the observations confirmed ultrasonographically. Heparinized blood samples were collected twice through jugular venipuncture seven days apart and the plasma progesterone concentrations measured by radio-immuno assay. Four buffaloes having smooth ovaries coupled with basal plasma progesterone ( $<1.0$  ng/ml) were classified as anoestrus and were excluded from the study. Twenty-seven buffaloes had a

CL and high plasma progesterone (>1.0 ng/ml), were classified as sub-oestrus and were subjected to Ovsynch treatment protocol.

The ovsynch treatment protocol was started irrespective of the stage of an oestrus cycle. On the first day (Day 0) 20 µg buserelin acetate (Receptal, 5 ml, Intervet, Holland) was injected intramuscularly, followed by 500 µg cloprostenol (Vetmate, 2 ml, Vetcare, India) on day 7 and another injection of 2.5 ml receptacle on day 9. Fixed time insemination was done 16 and 40 hours thereafter, using semen from progeny tested bulls of high genetic potential. Behavioural and clinical signs of induced oestrus, including bellowing, mucus discharge, tone in uterus and cervical status were recorded. Pregnancy was diagnosed by per-rectal examination, 90 days later. The results were subjected to Student's t-test.

### 3. RESULTS

#### 3.1 Factors affecting onset of puberty

Preliminary multiple regression analysis revealed that 7 out of 21 independent variables i.e. exercise of heifer, duration of colostrum feeding to calf per day, feeding of calf starter pre- and post-weaning, feeding till one year of age, type of calf starter and milk replacer and number of other growing calves in herd had no significant effect on the onset of puberty while the remaining 14 variables had significant collective effect ( $p < 0.01$ ). Regression analysis by stepwise deletion of these 14 variables revealed that factors such as education level and land holding of farmer, herd strength, wallowing, presence of bulls and, season of birth had no individual significant contribution on regression equation of age at puberty. The significance level of these variables varied from 0.072 (7.2%) to 0.964 (96.4%).

Regression analysis by stepwise deletion of variables considering their standard error revealed that occurrence of calf hood diseases was the most significant factor affecting the age at puberty in murrhah graded buffalo heifers of Punjab. This was followed by the feeding standards to heifers, occurrence of dystocia, bathing/wallowing practice during summer, suckling, housing index, influence of farm manager and the deworming status of the heifers. The occurrence of calf-hood diseases and of dystocia delayed the onset of puberty while better the feeding, bathing/wallowing during summer, provision of suckling, better housing index, experienced farm manager and deworming hastened its onset. The average age at puberty in buffalo heifers in different zones is given in Table I:

TABLE I. AVERAGE AGE AT PUBERTY OBSERVED IN BUFFALO HEIFERS OF DIFFERENT ZONES OF PUNJAB

Zone	Age at puberty (months) (Mean ±S.E.)	No. of heifers
Sub-mountainous	40.15 ± 0.83 <sup>a</sup>	217
Undulating plain	36.68 ± 0.56 <sup>b</sup>	216
Central plain	35.33 ± 0.56 <sup>b</sup>	247
Western plain	35.27 ± 0.48 <sup>b</sup>	243
Western region	35.61 ± 0.64 <sup>b</sup>	198
Flood plain	33.39 ± 0.68 <sup>c</sup>	218
Overall (Whole Punjab)	36.04 ± 0.68	1339

Values with different superscript vary significantly ( $P < 0.05$ )

Based on these, the final regression equation was derived:

Age at puberty (months) =  $55.33 + (15.547 \times \text{occurrence of calf-hood disease}) - (1.24 \times \text{feeding standard}) + (6.982 \times \text{occurrence of dystocia}) - (1.432 \times \text{bathing/wallowing practice during summer}) - (1.291 \times \text{suckling status}) - (0.562 \times \text{housing index}) - (0.632 \times \text{farm manager}) - (0.424 \times \text{deworming status})$

### **3.2 Effect of interventions on fertility in late pubertal buffalo heifers**

Four out of ten (40%) of the buffalo heifers supplemented with UMMB exhibited oestrus within one month and all conceived to the first natural service. All the heifers (100%) treated with eCG exhibited oestrus symptoms between 72-96 hours. However, none conceived to first service. Three of these heifers returned to oestrus between 10-21 days and got conceived (30% overall CR).

### **3.3 Success of OVSYNCH in the management of sub-oestrus**

Out of 220 buffaloes present at dairy farms, 31 (14%) were reported to be in anoestrus during winter months. However, clinical evaluation including ultrasonography and plasma progesterone analysis, confirmed true anestrus in only four buffaloes (13 %) whereas all the remaining 27 (87%) were sub-oestrus. Ovsynch treatment induced oestrus symptoms of varied intensity in all (100%). Of these, 18.51 % (5/27) buffaloes exhibited standing oestrus. Only 40.74% (11/27) buffaloes had vaginal mucus discharge with typical (77%) or atypical (23%) fern pattern. All (100%) buffaloes had variable degree of uterine tone, very good in ten, moderate in 11 and mild in six. Cervix was open enough for easy passage of AI gun in 89 % (24/27) but was dilated partially in the rest. All the animals had pink vestibule and swollen vulva.

Twenty-four out of 27 (89 %) buffaloes ovulated after ovsynch as confirmed by plasma progesterone conc.  $>1.0$  ng/ml 10 days after oestrus and 52% conceived to AI on the induced oestrus. The overall CR to three AI was 67%.

Investigations on the effect of size of follicle with vaginal discharge viz.-a-viz. CR revealed that all buffaloes with vaginal discharge had big follicle (Average  $8.5 \pm 0.94$  mm) at the time of first GnRH injection although two buffaloes with big follicle did not void any mucous. Buffaloes with mucus discharge at AI had better FSCR (73 %) than those without it. The intensity of uterine tone, however, did not appear to affect. Three out of 5 buffaloes showing overt signs of oestrus had large follicle ( $10.2 \pm 1.4$  mm).

Body condition score of the buffaloes affected their response to ovsynch. Four animals with relatively poorer BCS ( $2.58 \pm 0.33$  Vs  $2.98 \pm 0.21$ ) failed to resume cyclicity after induced oestrus. They also had milk production higher than the rest ( $5.00 \pm 0.91$  Kg. Vs  $2.42 \pm 0.80$  Kg.) which may account for their relatively lower BCS. Buffaloes with better BCS ( $2.87 \pm 0.13$ ) exhibited better signs of oestrus, which ultimately effected FSCR.

Size of follicle ( $6.91 \pm 0.69$  Vs  $6.05 \pm 0.9$  mm) or plasma progesterone conc. ( $1.43 \pm 0.15$  Vs  $0.76 \pm 0.12$  ng/ml) on day 0 of ovsynch had no bearing on the ovulation rate. Similarly, plasma progesterone conc. ( $0.85 \pm 0.14$  Vs  $1.91 \pm 0.23$  ng/ml) at the time of prostaglandin injection. had no relationship with ovulation rate. two buffaloes had pus flakes in vaginal mucus and did not conceive to AI at the induced oestrus.

## 4. DISCUSSION

### 4.1 Factors affecting onset of puberty

Of all the factors studied, puberty appeared to be affected most by the occurrence of calf hood diseases. Maximum age at puberty of 55.5 months was recorded in heifers which suffered from some or the other calf-hood disease while it was minimum (31.65 months) in heifers which remained healthy. Heifers dewormed regularly attained puberty significantly earlier than those without it (31.69 vs. 41.12 months,  $P < 0.01$ ), as has also been reported before [2].

Feeding standards of yearling heifers significantly influenced the onset of puberty. Well-fed heifers attained puberty much earlier than poorly fed (27.15 vs. 55 months). Calved allowed suckling matured faster than those weaned within a month (34.88 vs. 43.94 months). Heifers fed high-energy diet attained puberty 53 days earlier than moderate energy diet group [3].

Buffalo heifers given bathing once a day during summer months (April-September) attained puberty at the age of 42.55 months compared to 30.45 months in heifers who were allowed to wallow for more than 2 hours a day. Marked reduction in the age at puberty could be attributed to improved heat dissipation while bathing/wallowing [4,5]. Mean age at puberty of 38.49 months recorded in summer born calves was significantly higher than those born in any other seasons ( $P < 0.01$ )

Heifers reared under poor housing conditions required 55.61 months to reach puberty as compared to 32.22 months of heifers reared under excellent housing conditions ( $P < 0.01$ ). Houses protecting buffaloes from direct heat and with proper ventilation favoured efficient breeding [6,7] and faster growth in calves [8].

Calves delivered through dystocia ( $n = 142$ ) attained puberty at the mean age of 42.23 months compared to normally delivered calves ( $n = 1197$ ) at 35.31 months of age ( $P < 0.01$ ). Stress to pregnant animals may affect development of their foetuses [9].

Heifers having bulls in the herd ( $n = 803$ ) attained puberty at 33.31 months compared to 40.14 months in heifers without them ( $P < 0.01$ ) as has also been reported by [10]. Puberty was delayed (40 months) most in the submontaneous areas, probably because of lack of wallowing facility, poor housing and limited availability of green fodder, concentrates and minerals. Small quantities of local grasses and weeds formed the main part of the fodder. Poor housing causes physical discomfort and thermal stress. Further, improved housing together with better nutrition and mineral supplementation have been reported to hasten the onset of puberty in buffalo heifers [11]. Heifers receiving water splashing thrice a day conceived at an early age [12] corroborated our observations on the need for wallowing/bathing, especially during summer.

Earliest onset of puberty (33.39 months) was recorded in heifers reared around river basins of Punjab (Zone VI). Better availability of green grasses and fodders, together with wider practice of wallowing and supplementation of concentrates and minerals formed its merits. Within this zone, however, occurrence of calf hood diseases, dystocia and housing index significantly influenced the age at puberty in descending order. Decrease in age at first calving has been reported by timely adoption of prophylactic measures against contagious diseases like Hemorrhagic septicemia, Black Quarters and Foot and Mouth Disease [13]. Regression analysis for western plain zone (Zone IV) depicted that the education level of farmers on age at puberty contributed immensely. Active role of Multinational milk procurement agencies like Nestle India Ltd., played substantial role in educating and supporting the farmers with dairy production issues.

## **4.2 Effect of interventions on fertility in late pubertal buffalo heifers:**

### *4.2.1 UMMB Supplementation*

UMMB supplementation induced fertility in about 40% postpubertal anestrus buffaloes during summer. UMMB supplementation to anoestrus buffaloes during favourable breeding period (winter) has been reported to induce fertility in >80% [1]. Relatively poor results in the present study could be due to overriding effects of summer stress. The reproductive efficiency of buffaloes decreases during summer months presumably due to collective effect stress of heat, malnutrition and prolonged day length [14].

### *4.2.2 Hormonal Intervention*

Administration of eCG led to development of follicles and induction of oestrus in all treated animals. However, the conception remained very poor. eCG-induced stimulation of follicular growth failed to induce fertility in anoestrus buffaloes subjected to progestogen treatment [15].

## **4.3 Success of OVUSYNCH in the management of sub-oestrus**

The present study revealed that the incidence of true anestrus was low (1.8%) in winter while the rather high incidence of sub-oestrus (12.3%) was causing the same economic damage as true anestrus. The Ovsynch protocol, which narrows down the window of ovulation period thereby causing maximum synchrony between ovulation and insemination, proved successful in attaining 67% CR in sub-oestrus buffaloes. It has earlier been used for fertility management in cycling buffaloes [16]. The protocol also facilitated fixed time AI with semen from superior bulls and allowed genetic improvement. Size of follicle at the time of first GnRH injection, the BCS and uterine health etc. affected the success of ovsynch [16].

## **5. Transfer of technology**

After testing the efficiency of the Ovsynch protocol at two dairy farms, 11 field trials were facilitated for its wider application in different parts of Punjab state and some other states of India. An NGO, Dr A S Cheema Foundation Trust was involved to promote improved dairy cattle and buffalo production. More than 50 genetically superior females have already been produced (Photo). Information generated through this study was disseminated to working veterinarians, research workers, farmers and students through national seminars (n = 2), National Level Advanced training courses on reproduction (n = 2) and continuing Education Programme of this university (n = 5). International scientists visiting this university from Nepal, Myanmar and Mongolia were also trained in these techniques during the period under reference.

In conclusion, the study resulted in database on factors affecting the onset of puberty in buffaloes, standardized technology to enhance fertility in sub-oestrus buffaloes and promoted production of genetically superior buffalo heifers.

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# **INTEGRATION OF FEEDING STRATEGIES AND MANURE MANAGEMENT FOR IMPROVING GROWTH PERFORMANCE OF LOCAL CATTLE AND CONSERVING THE ENVIRONMENT UNDER FARMER'S CONDITIONS IN NORTH VIETNAM**

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## **Abstract**

Twelve farms with two lai sind cattle each were selected and divided into two groups to investigate effects of different feeding strategies on growth performance, feed utilization efficiency and methane production in cattle. In group I, animals were reared using the traditional feeding system (cattle fed untreated rice straw and King grass without urea molasses multi-nutrient block (UMMB) and in group II, animals were reared using a new feeding strategy (cattle fed urea-treated rice straw and UMMB). Coupled with the feeding trial, two manure management strategies (solid cattle manure managed in the traditional way without absorbent and stored in the field without cover vs. the new strategy were rice straw ash (15% w/w) is used as absorbent and manure is stored covered with plastic roof) were tested to determine the change in some of the major components i.e. dry matter, N and P in manure during 21 days of storage. After three months feeding trial, an agronomic trial using 700 m<sup>2</sup> plots divided into 12 blocks for the 4 treatments was conducted to investigate effects of the different fertilizers on biomass yield of King grasses and soil fertility. Results showed that new feeding strategy with urea treated rice straw and UMMB improved growth performance (15.6%), feed intake (12.4%) and feed conversion ratio (2.8%) of cattle and reduced methane production (17.7%) compared to that in traditional feeding system. Treatment of solid cattle manure with rice straw ash as an absorbent and keeping the manure covered with a plastic roof decreased of the leaching of N from manure by 75-77%. Applying the manure managed in new way to the soil improved soil fertility and biomass yield of King grass.

## **1. INTRODUCTION**

Mixed farming systems in which livestock rearing and crop production are the main components now are very common models of farming systems in Vietnam. For example, the pig rearing-rice production farm in Red river delta, rice production-fishery-duck rearing farm in Mecong river delta and Red river delta, local cattle rearing-rice and cassava production farm in the highland areas, etc. However, there are some challenges in these mixed farming systems. The biggest challenge facing the livestock sector is how to enhance animal productivity without adversely affecting the environment. In the local cattle rearing-rice and cassava production farming systems, productivity of the animals is restricted by low N and high fibre content of the native grasses and crop residues and their seasonal availability. Low quality, highly fibrous forages when fed to the cattle result in higher methane production compared with better quality forages. The production of methane in the rumen can represent a loss of 2-15% of the digestible energy depending on the type of diet [8]. Poor manure management also has been found to result in environmental pollution, low productivity of crops and soil degradation [6]. This study therefore focussed on identifying simple strategies that would decrease the production of enteric methane in local cattle, increase their growth performance and decrease loss of N and P into the environment from manure through appropriate manure management strategies.

## 2. MATERIALS AND METHODS

### 2.1. Location

The experiment was conducted at Anlao village, Vinh tuong district, Vinh Phuc province. Analyses were done at the laboratory of the National Institute of Animal Husbandry.

### 2.2. On-farm study

#### 2.2.1. Feeding trial and manure management

Twelve farms with two Lai sind cattle (18-20 month of age) each were selected and divided into two groups. In group I, animals were fed using the traditional system i.e. using untreated rice straw ad libitum with some elephant grass as the main source of fodder and 500 g of rice bran per head per day. Two manure management options were used in the trial. Solid manure was collected two times per day (morning and evening) and divided into two parts. The first part was managed using the traditional system i.e. fresh manure (solid) was collected and heaped on the road side, near the cattle shed or in the field without absorbent or covering (TM<sub>1</sub>) and the second part was stored in a hole covered with plastic roofing. Before storing in the hole, manure was mixed with rice straw ash (15%, w/w) as an absorbent and stored for three weeks before using it for as fertilizer (New manure management strategy; IM<sub>1</sub>). During storage, manure samples were collected at 0, 4, 14 and 21 days after treatment for analysis.

In the second group, animals in all households were reared by a new feeding strategy for hopefully both improving growth performance of the cattle and decreasing methane production. For this feeding strategy, animals were given ad libitum urea-treated rice straw [dry rice straw was prepared by spraying urea solution (4 kg urea dissolved in 100 liters water per 100 kg air dry straw) onto straw. While spraying the straw was thoroughly mixed and stored under airtight condition for two weeks] and urea molasses multi-nutrient block [UMMB : Sugar cane molasses (30%); rice bran (20%); urea (4%); cassava meal (23%); soybean meal (10%); bone meal (5%); Mineral premix (3%); salt (5%); bentonite (5%)] (500 g/head/day separated two times, morning and evening). Manure management applied for this group was the same as the first group with two kinds of manure TM<sub>2</sub> and IM<sub>2</sub>. Feeding trial was conducted for three months.

#### 2.2.2. Agronomical experiment

After three months of the feeding trial with two systems of manure management, an agronomical experiment was conducted to investigate effects of applying manure from different manure management strategies on biomass yield of king grass. A 700 m<sup>2</sup> plot was used for this study. The plot was planted with King grass and divided into 12 blocks in a completely randomized block design with 4 treatments (three blocks per treatment). The treatments were:

- (i) Treatment 1 : TM<sub>1</sub>
- (ii) Treatment 2 : IM<sub>1</sub>
- (iii) Treatment 3 : TM<sub>2</sub>
- (iv) Treatment 4 : IM<sub>2</sub>

The land was ploughed one month before planting and cattle manure (TM<sub>1</sub>; IM<sub>1</sub>; TM<sub>2</sub> and IM<sub>2</sub>) and was applied before planting at the rate of 1 kg (in DM)/m<sup>2</sup>. The first foliage was harvested 45 days after planting to estimate biomass yield. All the foliage was removed above 15 cm from ground level. Harvesting interval was every 45 days and after harvesting, cattle manure was applied at the rate of 0.5 kg (in DM)/ m<sup>2</sup> for all treatments.

### **2.3. Measurements**

Feed offered and refused were recorded daily to calculate feed intake and feed conversion ratio. Body weight of animals was weighed at every 2 weeks and linear regression analysis used to estimate rate of live weight gain. The manure from cattle (solid manure) in each household was collected and weight recorded daily for calculation of manure production. Daily methane production of cattle (litre of CH<sub>4</sub> per day) was estimated based on LIFE-SIM (Livestock Feeding Strategies-Simulation Model) software version 3.2 [11]. Manure sample from different management procedures were taken for analysis (mainly DM, N and P content) to investigate changes of these components by storing time.

Biomass yield of grass was measured at every harvesting and after two times of harvesting the soil samples of each treatment were taken to analyze for organic matter, N and P content.

### *2.4. Statistical Analysis*

Data were analyzed by analysis of variance using the general linear models (GLM) option of the ANOVA software of Minitab (Version 13.31) [12]. Differences among means were tested using Duncan's test.

## **3. RESULTS**

### **3.1 Growth performance, feed utilization, manure production and methane production of cattle in different feeding strategies.**

Growth performance, feed utilization efficiency, manure production and methane production of cattle reared in different strategies under smallholder's conditions are presented in Table I.

TABLE I. EFFECT OF FEEDING STRATEGIES ON FEED UTILIZATION EFFICIENCY, GROWTH PERFORMANCE, MANURE PRODUCTION AND METHANE PRODUCTION OF CATTLE UNDER HOUSEHOLD'S CONDITIONS

	Group I (TFS <sup>1</sup> )	Group II (IFS <sup>2</sup> )	SE
Number of animal	12	12	
Initial weight (kg)	179.8	181.1	6.84
ADG (g/day)	468.7 <sup>a</sup>	541.8 <sup>b</sup>	23.61
Dry matter intake (kg/day)			
Untreated rice straw	1.76	-	
Urea treated rice straw	-	1.86	
Elephant grass	1.84	1.83	0.11
Rice bran	0.43	0.43	0.022
UMMB	-	0.41	0.026
Total DM intake	4.03 <sup>a</sup>	4.53 <sup>b</sup>	0.24
Total DM intake as % BW	2.24 <sup>a</sup>	2.50 <sup>b</sup>	0.18
FCR (kg DM/ kg WG)	8.60 <sup>a</sup>	8.36 <sup>b</sup>	0.33
Manure production (kg/day)	7.65 <sup>a</sup>	8.01 <sup>b</sup>	0.29
Methane production (l/day)	0.16 <sup>a</sup>	0.13 <sup>b</sup>	0.006

<sup>1</sup>Traditional Feeding System;

<sup>2</sup>New Feeding Strategy

<sup>a,b</sup> Mean within rows with different superscripts differ significantly (P<0.05)

Total dry matter feed intake was different ( $P < 0.05$ ) and higher when cattle were given basal diets with urea treated rice straw and UMMB, but there were non significant different ( $P > 0.05$ ) on DM intake of elephant grass for cattle in both groups. Growth performance and feed conversion ratio of cattle in the new feeding strategy (group II) were significantly higher than those of traditional feeding system group. Manure production of cattle in group II was higher than that of group I about 4.7%. Methane production in cattle in group I was higher than that of group II about 17.7%. These data indicated that under smallholder's condition in the North Vietnam, feeding cattle with urea treated rice straw and UMMB (500 g per head per day) improved not only growth rate, feed utilization efficiency of animals, but also reduced methane production.

### 3.2 Change of component of manure during storing time

Changes of dry matter, N and P content in cattle manure managed in different ways in smallholder's conditions are showed in Table II.

TABLE II. EFFECT OF DIFFERENT MANAGEMENT WAYS ON CHANGES OF CHEMICAL COMPOSITION OF CATTLE SOLID MANURE UNDER SMALL HOLDERS' CONDITIONS

	TFS group		IFS group	
	TM <sub>1</sub>	IM <sub>1</sub>	TM <sub>2</sub>	IM <sub>2</sub>
<i>Day 0 (treatment day)</i>				
- DM (%)	19.2	22.6	20.7	23.01
- Nitrogen (% in DM basis)	1.74	1.52	2.16	1.88
- Phosphorus (% in DM basis)	0.36	0.41	0.34	0.38
<i>Seventh day after treatment</i>				
- DM (%)	24.6	23.7	23.9	24.3
- Nitrogen (% in DM basis)	1.22	1.38	1.47	1.71
- Phosphorus (% in DM basis)	0.35	0.39	0.33	0.38
<i>Fifteenth day after treatment</i>				
- DM (%)	30.5	25.1	32.6	27.4
- Nitrogen (% in DM basis)	0.78	1.31	0.99	1.64
- Phosphorus (% in DM basis)	0.34	0.37	0.32	0.35
<i>Twenty first day after treatment</i>				
- DM (%)	39.6	27.8	41.2	29.5
- Nitrogen (% in DM basis)	0.33	1.19	0.58	1.34
- Phosphorus (% in DM basis)	0.34	0.36	0.30	0.35

There were differences between cattle manure managed by traditional and new ways (TM<sub>1</sub> vs. IM<sub>1</sub> and TM<sub>2</sub> vs. IM<sub>2</sub>) in dry matter, N and P content. DM and P content in fresh manure (in treatment day) of cattle managed by new way were higher than that by traditional way, but N content in fresh manure managed by new way was little lower than that by traditional management. During storing there were big changes in DM and N content in the manure. DM content in manure managed by traditional management way (TM<sub>1</sub> and TM<sub>2</sub>) strongly increased (from 19.2% in day 0 to 39.6 % in TM<sub>1</sub> and from 20.7 % to 41.2 % in TM<sub>2</sub>) after 21 days of storing. In contrast, N content of cattle manure managed by this way markedly decreased (about 73 to 81%). With new management, DM content of manure increased about 23% (from 22-23% to 28-30%) and N content also decreased, but decrease level was much lower than those of traditional management way. Phosphorus content in cattle manure managed by new way was higher than that by traditional way, but it did not change much during storing time. Changes in DM, N and P content in the manure managed by different ways indicated that new strategy for manure management is a good way to prevent loss and escape of main nutrient (N and P) in manure to the environment.

### 3.3 Biomass yield of King grass

Biomass production of King grass fertilized with cattle manure treated by different ways is presented in Table III.

TABLE III: EFFECT ON BIOMASS YIELD OF KING GRASS FERTILIZED WITH CATTLE SOLID MANURE TREATED BY DIFFERENT PROCEDURES (TONES/HA)

	T <sub>1</sub> (TM <sub>1</sub> )	T <sub>2</sub> (IM <sub>1</sub> )	T <sub>3</sub> (TM <sub>2</sub> )	T <sub>4</sub> (IM <sub>2</sub> )	SE
<i>The first cutting</i>					
- Fresh matter	29.1 <sup>a</sup>	33.6 <sup>b</sup>	33.8 <sup>b</sup>	37.7 <sup>c</sup>	3.227
- Dry matter	4.83 <sup>a</sup>	5.78 <sup>b</sup>	5.89 <sup>b</sup>	6.48 <sup>c</sup>	0.321
<i>The second cutting</i>					
- Fresh matter	27.3 <sup>a</sup>	32.8 <sup>b</sup>	33.2 <sup>b</sup>	36.1 <sup>c</sup>	3.024
- Dry matter	4.58 <sup>a</sup>	5.54 <sup>b</sup>	5.66 <sup>b</sup>	6.17 <sup>c</sup>	0.298

<sup>a,b</sup> Mean within rows with different superscripts differ significantly (P<0.05)

There were significant differences in biomass yield between treatments (TM<sub>1</sub>, IM<sub>1</sub>, TM<sub>2</sub> and IM<sub>2</sub>) at the first and second cutting time. Biomass production at treatments that grasses were fertilized with cattle manure managed by new way was higher than that by traditional way at both cutting time.

### 3.4 Soil fertility

The results of soil characteristics in treatments are presented in Table IV. The data show a marked improvement in soil fertility as a result of fertilizing to grass with cattle manure treated by new way. Concentration of N (N), K (in K<sub>2</sub>O form) (K), P (in P<sub>2</sub>O<sub>5</sub> form) (P) were higher in the soil fertilized with cattle manure treated with absorbents and kept in the hole with plastic roof during storing time. There was a decrease of N, P, K concentration in the soil from the first to the second harvesting time.

TABLE IV: EFFECT OF FERTILIZING CATTLE SOLID MANURE TREATED IN DIFFERENT WAYS ON VARIATION OF SOLID COMPOSITION

	T <sub>1</sub> (TM <sub>1</sub> )	T <sub>2</sub> (IM <sub>1</sub> )	T <sub>3</sub> (TM <sub>2</sub> )	T <sub>4</sub> (IM <sub>2</sub> )
<i>At the first cutting</i>				
N	0.131	0.143	0.146	0.155
K <sub>2</sub> O	0.584	0.632	0.561	0.663
P <sub>2</sub> O <sub>5</sub>	0.091	0.102	0.088	0.104
<i>At the second cutting</i>				
N	0.102	0.133	0.118	0.136
K <sub>2</sub> O	0.432	0.516	0.455	0.531
P <sub>2</sub> O <sub>5</sub>	0.082	0.097	0.061	0.092

## 4. DISCUSSIONS

Under farmers' conditions in the Red river delta (RRD) of North Vietnam, commonly used feedstuffs for ruminants are crop residues. It is widely recognized that major limitations to the utilization of fibrous crop residues as a feedstuff for ruminants are associated with their low digestibility, low intake and low content of essential nutrients such as N and minerals. In this study improvement of growth performance of cattle in new group may be due to contribution of the urea treated rice straw and UMMB. Many reports show that treatment of rice straw with urea and other chemicals improved nutritive value of rice straw. Vu Duy Giang [9] reported that treatment of rice straw with urea and Ca(OH)<sub>2</sub> improved organic matter digestibility in cattle compared to untreated rice straw and concentration of NDF tended to reduce and voluntary feed intake in cattle

increased when they were fed urea treated rice straw. Other workers also reported positive responses of cattle in growth performance and feed utilization efficiency when they were given urea treated rice straw compared to un treated rice straw such as [5, 10]. In this study, one of reasons for improving performance of the animals in the new group is addition of UMMB to diet for cattle. UMMB supplied cattle with some essential nutrients such as by-pass protein and minerals. So with supplementation of UMMB to the ration together with urea treated rice straw improved cattle performance. These results agree with results in research of Badudeen et al., [1] and Ghebrehiwet et al., [2].

Methane production in animals during the process of fermentative digestion and metabolism represents a loss of food energy. It was observed that methanogenic energy loss is high particularly on low quality roughages such as straw-based diet [3]. In this study, lower methane production was found in the animals at the new feeding strategy group. This may due to quality of roughages for animals in these groups was improved by treatment of rice straw with urea and due to contribution of UMMB supplementation. Leng [4] reported that 2 kg of methane produced per kg meat when animals fed mainly with straw. However, methane production can be reduce to 0.36 kg to produce kg meat by offering straw with urea and by-pass protein which may be due to the efficient fermentative digestion. The results of this study agree with statement of Saadullah [7] that the methane production per unit of product can be reduced by 25 to 75% allowing urea molasses block, fishmeal, oil cakes etc.

In rural areas in RRD in Vietnam, farmyard manure commonly applied to the soil when it is fresh or it can be stored. Fresh manure contains large amount of easy available nutrients, but these nutrients may be easier lost under poor management. The data in Table 2 indicate that with traditional management way N loss is highest during storing time and it is a main reason for low biomass production of grasses fertilized with manure treated in traditional way.

## 5. CONCLUSION

The new feeding strategy improved growth performance, feed utilization efficiency and reduced methane production in local cattle. Applying to the soil solid cattle manure absorbed with rice straw ash and stored in the hole with plastic roof for three weeks before planting improved biomass yield of elephant grass and soil fertility.

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# INTEGRATION OF FEEDING STRATEGIES AND MANURE MANAGEMENT FOR IMPROVING MILK PRODUCTION OF DAIRY COWS AND CONSERVING THE ENVIRONMENT UNDER FARMER'S CONDITIONS IN SOUTH VIETNAM

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## Abstract

Six farms with 4 dairy cows (75% of Holstein Friesian) each in Hoc Mon district, Ho Chi Minh City, Vietnam were selected and divided into two groups (three farms; 12 cows each). In group 1, animals were fed using traditional feeding system versus a new feeding strategy in group 2. The diet in the traditional feeding system was composed of grasses, untreated rice straws, cassava waste, brewery waste and concentrate. In the new feeding system, rice straw was treated with 4% urea and animals were supplemented with urea molasses multi-nutrient blocks (UMMB). In each group, the animals were further divided into two groups for manure collection. In one group, the traditional manure storage method i.e. sun-drying was used whereas in the other, manure was stored roofed and covered and the manure from the different storage methods subsequently used as fertilizer for grasses. The results showed that the new feeding system with urea treated rice straw and UMMB improved milk production and reduced methane production. Roofing and covering manure reduced N loss from the manure and improved soil fertility and biomass yield of the test grasses.

## 1. INTRODUCTION

Livestock production is a traditional sector, which is developing rapidly in Vietnam. The population of ruminants in 2007 was 6.8 million heads of cattle, 3.0 million buffaloes and 1.8 million goats and sheep. Ruminant production in Vietnam is different from those in developed countries in that it is mainly composed of small farms with two to 4 heads of cattle/buffaloes and/or 10 to 15 sheep and goats. Coupled with the small farm sizes is low knowledge and skill of farmer implying most livestock is reared in a semi-intensive systems, which combines grazing, and supplementation of crop residues such as rice straw and peanut hay at cattle sheds after grazing.

Most of dairy farms are concentrated around Hochiminh City and Hanoi the capital for close proximity to the milk processing factories. Currently, the dairy cattle population is approximately 100,000 with almost 70,000 of the cattle around Hochiminh City. As the land size for dairy farmers is gets smaller and smaller and the increasing population of dairy cattle around the cities, the issue of roughage shortage and environmental pollution becomes of great concern.

Furthermore, as a hot and humid tropical country, the quality of forages and crop residues is low. This leads to low animal productivity and high methane production from the animals due to inefficient fermentation in the rumen. Low quality roughage and the nutritionally imbalanced diets, especially energy and protein are the main factors effecting methane production. Thus, treating of roughages with urea, feed supplementation, balancing the ration and improving the environmental condition of

cattle sheds are basic methods to decrease methane production and improve the efficiency of feed utilization and therefore increase the overall productivity of the animals.

Additionally, because Vietnam is an agriculture country, people have always used manure for crop production. However, the management and utilization of manure are still very simple resulting in considerable losses of N and P into the environment. The traditional methods are composting manure with others waste such as straw, grasses, tree leaves. For a faster composting process, farmers normally add lime to the compost and this causes more losses of nutrients from the manure. Another method for treatment manure is sun-drying, but it also leads to nutrient losses especially N. Recently, farmers have started making manure compost with yeast or using red worms (*perionyx excavatus*) to treat the manure which shorten the time required to cure the compost as well as to reduce the loss of nutrients. In small pig or dairy farms, biogas production is also normally used for manure treatment. Solid waste, urine and wastewater from stable are gathered into plastic bags or concrete pool for biogas production. The waste product after fermentation has a high nutrient value and is used to fertilize grasses.

The overall objective of this project was to improve livestock productivity through better nutritional strategies while conserving the environment. The specific objectives were to improve milk production and decrease methane, N and P pollution into the environment in dairy farms.

## 2. MATERIALS AND METHODS

### 2.1 Location, number of farms, animals and grassland area

The Project was conducted at Dong Thanh village, Hoc Mon district, Ho Chi Minh City (about 15 km from the city centre). Six dairy farms with around 12 heads of cattle of which 4 cows were milking (2<sup>nd</sup> to 3<sup>rd</sup> month of lactation) were chosen for the feeding and manure management strategies study. Each farm had at least 2,000 m<sup>2</sup> of grassland

### 2.2 Experimental design

#### 2.2.1 *On-farm feeding and manure storage strategy*

The 6 farms were divided into two groups i.e. three farms with 12 dairy cows per group. In group 1, animals were fed using the traditional system whereas in group 2, a new feeding strategy was used. In the traditional feeding system (TF), cows were fed 20 to 25 kg of grasses plus 3 to 5 kg of untreated rice straws, 4 to 6 kg of cassava waste and 6 to 8 kg of brewery waste was as basal diet. The amount of concentrate fed was based on milk yield i.e. 0.4 kg of concentrate for 1 kg of produced milk. In the new feeding system (NF), cows were fed grasses, cassava waste and brewery waste as above with ad libitum urea (4%) treated rice straw and supplemented with UMMB. The amount of concentrate fed was based on milk yield as above but at 0.3 kg of concentrate per kg of milk produced. The rice straw was treated spraying dry straw with urea solution (4 kg urea dissolved in 100 litres of water for every 100 kg air-dry straw). After spraying, the straw was thoroughly mixed and stored under airtight condition for 2 weeks. The UMMB was composed of sugar cane molasses (40%); rice bran (40%); urea (5%); mineral premix (1%); salt (1%); Lime (8%); Cement (5%).

The animals were further divided into two sub-groups for manure collection. Manure from one sub-group was stored using the traditional method (sun-drying) and the other using a new method i.e. by roofing and covering. In the traditional manure storage

method, fresh solid manure was collected and piled on the road or yard, near the cattle shed or in the grassland without any absorbent and cover whereas for the new manure storage method, fresh solid manure was collected and piled in a brick-pool with roofing and plastic covering to protect from sun light and wind. Rice hull (10%) was used as an absorbent material. Manure from the different storage methods were subsequently used as fertilizer in an agronomic study, which lasted 120 days. The manure was stored

### 2.2.2 Agronomic manure evaluation

Elephant grass (*Penisetum purpurium*) in the 2<sup>nd</sup> year after planting exists in all farms. A plot of 900 m<sup>2</sup> was used in the study. The plot was divided into 6 blocks (150 m<sup>2</sup>/block) in a completely randomized block design with two treatments and three blocks per treatment. Chemical fertilizer was not used on the plots although manure had been used (2,000 kg/ha/cutting at day three after cutting. The cutting interval was 45 days) on all plots. Manure was composted for 2 months before using as fertilizer. The trial lasted 180 days (4 cutting).

## 2.3 Measurements

- a) Daily measurement of feed intake to calculate DM intake. The feed were sampled daily and composited weekly for compositional analysis (DM, CP, CF, NDF, ADF) in triplicate.
- b) Milk production was measured daily and sub-sampled for compositional (DM, Fat, Protein) analysis.
- c) Methane production (liter of CH<sub>4</sub> per animal per day) was estimated based on LIFE-SIM (Livestock Feeding Strategies-Simulation Model) software version 3.2 developed by ILRI (International Livestock Research Institute, 2003).
- d) Solid manure production was measured daily and sub-sampled in triplicate on day 0, 7, 15 and 21 for compositional analysis (DM, Nitrogen and Phosphorus).
- e) Biomass yield of grass (Fresh and dry matter) was determined after each cut for each of the 4 cut.
- f) The soil (1 sample per block) was also sampled before and after experiment and after each of the 4 cut for nutrient composition (N, K<sub>2</sub>O, P<sub>2</sub>O<sub>5</sub>) determination.

## 2.4 Statistical Analysis

Data were analyzed by analysis of variance using the general linear models (GLM) in Minitab (Version 13.31) 2000. Differences among means were separated using Duncan's new multiple range test (MRT).

## 3. RESULTS AND DISCUSSION

### 3.1 Effects of new feeding system on feed intake, milk production, manure production and methane production of dairy cows

#### 3.1.1 Chemical composition of feedstuffs

The chemical composition of feedstuffs used in the diets of dairy cows is showed in Table II.

TABLE II. CHEMICAL COMPOSITION OF FEEDSTUFFS

Feedstuffs	DM (%)	CP (% of DM)	CF (% of DM)	NDF (% of DM)	ADF (% of DM)
Grass	15.6	12.1	30.2	64.2	32.1
Rice straw	91.8	4.6	32.6	67.3	40.1
UTRS*	49.1	8.6	24.5	63.1	35.8
Cassava waste	17.2	2.6	8.4	19.2	10.4
Brewery waste	22.6	31.7	13.9	57.9	23.6
Concentrate	90.1	18.2	5.4	52.7	24.6

\*UTRS – urea treated rice straw

Data on Table II showed that most of the feedstuffs in dairy cattle diets have a high content of water So that dry matter of feedstuffs is low: Grass 15.6%, cassava waste 17.2%, brewery waste 22.6%. On the other hand, untreated rice straw and cassava waste have a low content of protein: 4.6% and 2.6%. Grass and untreated rice straw have a high content of fibre: 30.2% and 32.6%.

There were many studies on treatment of feedstuffs, especially fibre by-products in other to increase nutritive values. Doan Duc Vu *et al.*, 1999<sup>a</sup>; Doan Duc Vu *et al.*, 1999<sup>b</sup>; Doan Duc Vu, 2000; Nguyen Xuan Trach, 1998; Nguyen Van Thu, 1997; Vu Van Noi *et al.*, 2001 have studied on treatment of rice straw with urea. Bui Van Chinh, 2001; Nguyen Thi Tu, 2001 have results of treatment of sugarcane leaves and bagasses. Bui Xuan An, 1998; Doan Duc Vu, 2004 have studied on peanut vines and pineapple waste silages. It concluded that the fibre by-products can be improved with some treatment methods.

### 3.1.2 Effects of new feeding system on feed intake of dairy cows

The quantity of feedstuffs and nutritive values of diets are showed in Table III. According to NRC, nutrient requirement of a dairy cow with 450 kg of body weight and 16 kg of milk yield is 13.10 kg DM, 2.80 kg CF, 1.80 kg CP and 33.55 Mcal ME. Data of Table III showed that a dairy cow could intake 23.4 kg grasses, 1.8 kg of untreated rice straw, 4.8 kg of cassava waste, 6.8 kg of brewery waste and 6.2 kg of concentrate. Daily DM intake and energy was not enough compared to requirement while fibre content was high but protein content was low. When rice straw has been treated with urea and UMMB was supplemented, the nutrient value of the diet has been improved. All of nutrient indices including DM, CF, CP and ME have been met compared requirement.

TABLE III. FEED INTAKE OF DAIRY COWS FED DIFFERENT DIETS

Indices	Group 1	Group 2
Feed intake (kg/day)		
- Grasses	23.4 ± 4.6	22.5 ± 2.7
- Untreated Rice straw	1.8 ± 0.9	-
- Urea Treated Rice Straw	-	7.6 ± 0.3
- Cassava waste	4.8 ± 1.7	4.2 ± 1.5
- Brewery waste	6.8 ± 2.0	7.3 ± 1.6
- Concentrate	6.2 ± 0.9	4.8 ± 0.2
- UMMB	-	0.62 ± 0.1
Nutrient value of diet (kg/day)		
- DM	11.18 <sup>a</sup> ± 0.7	13.12 <sup>b</sup> ± 0.4
- CF	2.36 ± 0.18	2.57 ± 0.19
- CP	1.47 ± 0.14	1.68 ± 0.11
- ME, Mcal/day	31.17 <sup>a</sup> ± 2.30	33.44 <sup>b</sup> ± 1.73

<sup>a,b</sup> Mean within rows with different superscripts differ significantly (P<0.05)

There are some methods for improving dairy cattle diets in Vietnam condition. Under project RAS/5/030, RAS/5/035 and other national and international projects, UMMB technology was widely disseminated and adopted by farmers in many regions in the whole country (Doan Duc Vu *et al.*, 1999; Nguyen Van Thu *et al.*, 1993; Bui Xuan An *et al.*, 1993; Bui Van Chinh *et al.*, 1993). With crude protein around 25%, cassava hay fed with 2kg/herd/day helps to balance the energy and protein ratio in dairy cattle diets. This protein stimulates both productivity and efficiency of roughage utilization (Doan Duc Vu, 2003). Supplementation of whole cottonseed or cottonseed meal with amount of 2 kg resulted to have a long peak of lactation of dairy cows (Dinh Van Cai, 1999).

### 3.1.3 Effects of new feeding system milk production, manure production and methane production of dairy cows

The milk production, milk composition, manure production and methane production are showed in Table IV. Data of Table IV showed that, with the new feeding system (urea treated rice straw + UMMB) milk production of dairy cows increased considerable ( $P<0.05$ ) compared to control group: 16.27 vs. 15.01 kg/cow/day. Milk composition (DM, fat and protein) also has been improved but differences of data between two groups were not significant. The costs of feedstuffs for one kg of produced milk in new group was lower 0.06 USD/kg than that in traditional feeding group (0.19 vs. 0.25 USD/kg). The amount of produced manure of dairy cows in group two was higher than that in group 1 ( $P<0.05$ ): 24.5 vs. 21.6 kg/cow/day while methane production of dairy cows decreased when the animal diet has been intervened.

TABLE IV. MILK PRODUCTION, MANURE PRODUCTION AND METHANE PRODUCTION OF DAIRY COWS FED DIFFERENT DIETS

No.	Indices	Unit	Group 1	Group 2
01	Number of dairy cows	Head	12	12
02	Milk production	Kg/cow/day	15,01 <sup>a</sup> ±0,68	16,27 <sup>b</sup> ±0,72
03	Milk composition: - DM	%	11,38±0,54	11,99±0,54
	- Fat	%	3,38±0,45	3,75±0,44
	- Protein	%	3,17±0,18	3,46±0,15
04	Feed cost for 1kg of milk	USD/kg	0.25	0.19
05	Manure production	Kg/cow/day	21.6 <sup>a</sup>	24.5 <sup>b</sup>
06	Methane production	L/cow/day	0.42 <sup>a</sup>	0.33 <sup>b</sup>

<sup>a,b</sup> Mean within rows with different superscripts differ significantly ( $P<0.05$ )

Milk production has been improved due to using urea treated rice straw, UMMB leading a suitable rumen environment for activities of microorganisms. Many authors have proved these results such as Doan Duc Vu 2000, Bui Van Chinh and le Trong Lap, 1996. Methane is produced during the digestion in the rumen of ruminants. Low quality roughage, the nutritionally imbalanced diets, especially energy and protein are main factors effecting methane production. Thus, treating of roughages, feed supplementation, balancing the ration and improving the environmental condition of cattle sheds are basic methods to decrease methane production, to improve efficiency of feed utilization and to increase productivity of ruminants.

In the previous time, the measurement of methane production from livestock has not been considered. However, yet in two recent years, within the framework of cooperation with International Livestock Research Institute (ILRI), we tried to use simulation model software, which can determine the methane production on each

individual animal, on each kg of milk or meat produced. This software can help estimating the changing of methane production between different sources of feedstuff and those from different rearing methods.

The appropriate strategies for reduction of methane production are still mainly based on feeding strategies, in which treatment of roughage, use of feed supplementation, balancing the ration could be applied to the farm household. Saadullah, 1981 concluded that while 0.126 kg methane produced per kg live weight gain with urea treated straw, it was 0.84 kg per kg live weight gain with untreated straw. Saadullah, 1992 proved that the methane production per unit of product can be reduced by 25% to 75% allowing urea molasses block, fish meal, oil cakes etc. Ionophore feed additives (monensin sodium) supplementation into concentrate feeds to promote propionate and reduce methane production by 16-24% in steers (Thromton and Owens, 1981).

### 3.2 Effects of new manure management on manure quality, grass yield and soil quality of grassland

#### 3.2.1 Effects of new manure management on manure quality

The chemical composition of cattle solid manure with different methods of storage is showed in Table V.

TABLE V. CHEMICAL COMPOSITION OF CATTLE SOLID MANURE WITH DIFFERENT METHODS OF STORAGE

No.	Indices	Unit	Group 1		Group 2	
			Sub-group 1 (TF-TM)	Sub-group 2 (TF-IM)	Sub-group 3 (IF-TM)	Sub-group 4 (IF-IM)
Day 0						
01	DM	%	19.8	27.3	20.7	28.6
02	Nitrogen	%/DM	1.86	1.62	2.14	1.82
03	Phosphorus	%/DM	0.33	0.46	0.31	0.48
7 <sup>th</sup> day						
04	DM	%	25.6	24.7	26.9	26.3
05	Nitrogen	%/DM	1.52	1.58	1.67	1.71
06	Phosphorus	%/DM	0.34	0.36	0.31	0.32
15 <sup>th</sup> day						
07	DM	%	32.5	25.8	32.8	25.4
08	Nitrogen	%/DM	0.76	1.43	0.59	1.65
09	Phosphorus	%/DM	0.44	0.38	0.42	0.39
21 <sup>st</sup> day						
10	DM	%	44.6	27.2	47.2	28.5
11	Nitrogen	%/DM	0.45	1.39	0.48	1.45
12	Phosphorus	%/DM	0.41	0.46	0.35	0.33

There were differences in dry matter, N and P content of cattle manure between traditional and new methods of manure storage. In general, P almost has not been changed while N decreased in traditional group compared to new group. There were not differences in indices between traditional and new feeding systems.

Vietnam is an agriculture country so people have used manure for crop production for a long time. However, the management and utilization of manure are still very simple inducing the losses of N and P into the environment. Under the project funded by SAREC/SIDA, some of works have been done to evaluate efficacy of utilization of chicken manure and chicken litters as feeds for animals. Tran Quoc Viet *et al.*, 2001

reported that rice straw chicken litter (RSCL) could be a valuable feed resource for local cattle in dry season in North Vietnam. However, in order to get high feeding efficiency, rice straw chicken litter should not be used as a sole feed for cattle. The best method is combination rice straw chicken litter with urea-treated rice straw (UTRS) at the ratio of 30% (RSCL) and 70% (UTRS) (w/w). Ninh Thi Len et al (2003) reported that chicken manure could be ensiled for at least 90 days with mixtures of cassava root meal, rice bran, sugar cane molasses and cassava residue, and the final product used as feed resource for fattening pigs.

### 3.2.2 Effects of new manure management on grass yield fertilized manure

The biomass of grasses fertilized manure with different methods of storage is showed in Table VI. Data of Table VI showed that when stored cattle manure has been used for fertilizing grasses, the production of grasses increased ( $P < 0.05$ ) compared to using untreated manure: 46.4 vs. 51.4 Ton/ha/cutting in traditional feeding system group and 42.2 vs. 43.1 Ton/ha/cutting in new feeding system group. Results of dry matter of grasses were the same in fresh matter.

TABLE VI. BIOMASS OF GRASSES FERTILIZED MANURE WITH DIFFERENT WAYS OF STORAGE

No.	Indices	Unit	Group 1		Group 2	
			Sub-group 1 (TF-TM)	Sub-group 2 (TF-IM)	Sub-group 3 (IF-TM)	Sub-group 4 (IF-IM)
01	# of cutting	Times	4	4	4	4
02	Fresh matter	Ton/ha/cutting	42.2 <sup>a</sup>	48.4 <sup>b</sup>	43.1 <sup>a</sup>	51.4 <sup>b</sup>
03	Dry matter	Ton/ha/cutting	6.38 <sup>a</sup>	7.12 <sup>b</sup>	6.46 <sup>a</sup>	7.67 <sup>b</sup>

<sup>a,b</sup> Mean within rows with different superscripts differ significantly ( $P < 0.05$ )

### 3.2.3 Effects of new manure management on soil quality of grassland fertilized manure

The chemical composition of soil fertilized manure with different methods of storage is showed in Table VII. Data of Table VII showed that after some harvests of grasses, concentration of N, K (in  $K_2O$  form) and P (in  $P_2O_5$  form) in the soil fertilized with stored cattle manure was higher than that with untreated manure.

TABLE VII. CHEMICAL COMPOSITION OF SOIL FERTILIZED MANURE WITH DIFFERENT METHODS OF STORAGE

No.	Indices	Unit	Group 1		Group 2	
			Sub-group 1 (TF-TM)	Sub-group 2 (TF-IM)	Sub-group 3 (IF-TM)	Sub-group 4 (IF-IM)
Before experiment						
01	N	%	0.16	0.14	0.13	0.15
02	$K_2O$	%	0.45	0.63	0.56	0.66
03	$P_2O_5$	%	0.08	0.10	0.08	0.10
After experiment						
04	N	%	0.10	0.18	0.11	0.19
05	$K_2O$	%	0.33	0.71	0.34	0.87
06	$P_2O_5$	%	0.06	0.11	0.07	0.13

In Vietnam, cattle manure commonly applied to the soil in fresh or stored forms. Fresh manure contains large amount of easy available nutrients, but these nutrients may



be easier lost under poor management. The data in Table V indicate that with traditional method N loss is highest during storing time and it is a main reason for low biomass production of grasses fertilized with manure treated in traditional way. Parallel this, the quality of soil fertilized with untreated manure was lower than that new managed manure.

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# **INTEGRATED APPROACH IN IMPROVING LIVESTOCK-CROP FARMING USING INDIGENOUS RESOURCES AND CONSERVING THE ENVIRONMENT**

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## **Abstract**

On station trials were conducted to evaluate the effects of cattle feed composition on the quality of manure and the effects of manure storage systems on crop yield. Statistical analysis showed that there were significant increases in soil organic matter and soil porosity in manured plots over the control. In the second trial, porosity decreased due to the increase in bulk density. The results also showed that there were increases in the soil aggregate stability in the manured plots but the increases were not significant over the non-manured plots. The first experiment showed that dry matter yield of plants in the manured-plots and in plots fertilized with inorganic fertilizer was not significantly higher than the non fertilized control treatment. No significant increases were obtained in the grain yield of corn with the treatments imposed beyond that achieved by the non-fertilized control during the first experiment. In the second experiment (residual), the same trends were observed in dry matter yield as in the first experiment with non significant increases in the dry matter yield in treated plots. In sub-plot b1 (with supplemental N) application of manure with improved manure management resulted in the highest N yield in stover of the treatments measured. Results obtained from inorganic fertilizer application and conventional storage were comparable. In corn grain, the treatment receiving organic fertilizers alone had the highest N yields but was not significantly different from the manure treatment with improved manure management or conventional manure storage. A similar trend was observed for Ndff, FN yield and Fertilizer Nitrogen Use Efficiency. The highest plant N-yield was obtained in the treatment with best feeding and manure management practices (T6) although the N-yield was not significantly higher than for the other treatments. Similar trends in yield were observed in the first and second experiments. Using the isotope technique, nitrogen derived from fertilizer (Ndff), fertilizer N Yield (FNY) and Fertilizer Nitrogen Use efficiency were calculated using sub-plots b2. The treatment with improved nutrition and manure management provided fertilizers-N for the first crop with the highest % Ndff (Nitrogen derived from fertilizer) and Fertilizer Nitrogen Yield (FNY) in the grains and stover but was not significantly different to that from inorganic fertilizers N source and conventional manure storage. The Ndff for the inorganic fertilizer treatment was slightly higher in the roots compared to manure treatments with conventional and improved manure management in the second crop to measure residual fertilizer effects.

## **1. INTRODUCTION**

The Philippine dairy sector has a very low productivity due to poor farm and nutritional management. The productivity of this agricultural sector will need to improve to reduce the cost of imports in the face of rising global food prices [1].

The increasing demand for food with the associated rise in global population has led to elevated demand for scarce fertilizers to maintain crop production. However, the current energy crisis coupled with the rise in cost of raw materials and labour has led to increased prices and reduced production of inorganic fertilizers. In the current environment, there is a need to consider substitutes and/or supplements to the use of organic fertilizers.

Animal manure is the most basic agricultural by-product and has a long history as a fertilizer in agriculture. However, to activate the benefits of manure fertilizers there is a need to manage this fertilizer input to prevent environmental problems with its use. Excess manure application or poor storage of manures can pollute the environment. To be a sustainable fertilizer source, livestock and/or crop farmers should undertake management practices that minimize the negative impacts of manures on the environment. Manure management and utilization is therefore an important component of a sustainable livestock and/or crop farming systems in order to optimize the use of livestock manures as organic fertilizers for cropping and pastures.

This study was undertaken to evaluate the effects of feed composition on manure quality and manure storage systems on crop yield.

## 2. MATERIALS AND METHODS

### 2.1 Feeding management of dairy cows used as manure sources

#### 2.1.1 Basal ration group

Dairy cattle were only allowed limited grazing time during the day with cut and carry feeding in the morning and afternoons from pasture and/or native plant resources, consisting mainly of grasses (e.g. *Paspalum auritum*, *Paspalum conjugatum*, *Brachiaria mutica* and *Pennisetum purpureum*) and mixed forage (principally legumes such as *Centrosema* spp. and *Gliricidia* spp.). Agricultural by-products when they are available such as rice straw are also provided during the drier months of March to May. Daily dry matter intake of roughage for these farms was calculated to be 8 kg/cow. A lactating dairy cow concentrate (Crude protein=16%, crude fat=4%) was also provided at the rate of 3-4 kg/head/day consisting of corn, corn grits, sorghum, soybean oil meal, rice bran, wheat bran, corn bran, pollard, molasses, corn gluten, ipil-ipil meal, copra meal, corn germ meal, brewer's yeast, limestone, salt, dicalcium phosphate, vitamins, trace minerals, antioxidant and mould inhibitor. The percentage of each ingredient was not provided in the accompanying literature from the concentrate manufacture.

#### 2.1.2 Block supplemented group

In addition to the basal diet, a group of dairy cows were fed 500 grams/UMMB (urea-molasses multinutrient block) per day. The block was cut in half with one half being fed in the morning and the other half in the afternoon. Composition of UMMB was (%); molasses, 40; rice bran, 40; urea, 8; cement, 10; salt, 1.8, mineral premix, 0.2. However, the percentage of molasses and rice bran in the blocks varied according to the consistency of the molasses.

## **2.2 Manure management**

### *2.2.1 Conventional management*

Before the cows were milked, manure was manually removed from the animal housing to an uncovered manure pile. Once the solid waste was removed, the floor was cleaned using a continuous flow of water from a hose.

### *2.2.2 Improved manure management*

The manure storage area was covered with a roof and the manure surface covered with a very thin layer of soil approximately two to three days interval.

## **2.3 Manure evaluation/utilization study**

Manures from two groups of cattle, one group (n = 8) was fed the basal ration and other group (n=8) was fed the basal ration supplemented with UMMB, collected, manures from each subdivided so there were two manure piles for each feeding group and stored from March to May 2006 (average temperature was 27.9 °C; rainfall for the period was 122.8 mm). The manures from each feeding group that had been subdivided were stored as either conventional and improved manure management as described above. These manures were produced and stored at Sariaya, Quezon, approximately 120 kilometers south of Manila (Latitude 13° 58' N, Longitude 121° 31' E). The manures were transported to the Bureau of Soils and Water Management National Soil and Water Resources Research Center (NSWRRC) for further evaluation and their efficiency as fertilizers.

The dried manures were subsamples and analyzed for nitrogen content by Kjeldahl method and the P,K, Ca, Mg, Zn, Cu, Fe and Mn contents (Table 1.) were determined by wet ashing methods from vol. 1.1988 ed. of the Methods of Soil, Plant, Water and Fertilizer Analysis for Research of the Bureau of Soils and Water Management [2].

The fertilizer plots were located at the NSWRRC site, San Ildefonso, Bulacan (Latitude 15° 4' N, Longitude: 120° 56' E), approximately 66 kilometers north of Manila. The soil in the experimental area is classified as a Vertisol (Typic Haplusterts) on an alluvial plain with a slope of less than 1%. These soils are mainly utilized for wetland rice production during the wet season with corn and grain legumes being grown during the dry season.

Prior to the fertilizer experiment, the area was cleared of all debris and composite soil samples were collected for chemical and physical analyses, which are presented in Table II. Soil organic matter was determined by Walkley-Black Method. Cored soil samples of the topsoil were also collected to determine soil bulk density analysis according to the method of the Bureau of Soils and Water Management.

The first fertilizer experiment was carried out from January to March, 2007 with an average daily temperature of 26.9°C and 16 mm of rainfall for the period. The second experiment to determine the residual fertilizer value of the manures was carried out from August to November 2007 with an average daily of 27.3°C and 248 mm of rainfall for the period. Corn cv. IPB 929 was used as the test crop to evaluate the fertilizer responses for both experiments.

The fertilizer treatments for the main and subplots applied to the soils in the experiment were as follows:

**Main Plot: (Manure type/storage method)**

T1- control (no manure; no inorganic fertilizer)

T2- Inorganic fertilizer (120-30-30 kg NPK/ha)

T3- Manure from conventionally-fed cattle with conventional storage

T4- Manure from conventionally-fed cattle with improved manure management

T5- Manure from UMMB-fed cattle with conventional storage

T6- manure from UMMB-fed cattle with improved manure management

**Sub-plots: (N side dressing as ammonium sulphate)**

A. no side-dressing of N

B. with side dressing of 40 kg N/ha at 30 days post planting and 40 kg/ha N at silking stage).

The treatments were laid-out in a split-plot randomized design with the manure type/storage method as the main plots and side dressing of N as the sub-plots. Each treatment was replicated three times. The layout of plots is shown below.

Replication	Treatment number/Subplot A or B											
1	3A	3B	4A	4B	1A	1B	2A	2B	5A	5B	6A	6B
2	6A	6B	1A	1B	5A	5B	3A	3B	4A	4B	2A	2B
3	1A	1B	2A	2B	5A	5B	4A	4B	6A	6B	3A	3B

*FIG 1. Treatments*

The main plots were 8 × 5 meters with sub-plot of 4 × 5 meters covering one-half of the main treatment plot and the sub-plots were randomized accordingly with the main plots. Two weeks prior to planting, the corn seed cattle manure was applied to the designated plots at the rate of 7.5 tons/hectare.

**2.4 Establishment of <sup>15</sup>N sub-plots and <sup>15</sup>N isotope labelling of the soil**

Three sub-plots with an area of 0.64 m<sup>2</sup> (80 cm x 80 cm) were established in treatment plots 2, 3 and 6 using marine ply boxes half buried in the soil to contain the lateral spread of the applied treatments in all the three replicates. Two micro-plots were established with the boxes in the N-top-dressed subplot and one in the non top-dressed half of the main plot. These sub-plots were designated as a, b1 and b2 as shown in Figure 2. <sup>15</sup>N-labelled ammonium sulphate with 10.27% N atom excess was applied to the soil inside sub-plots a and b1 at the rate of 20 kg/ha 2 weeks before planting and at seeding. The soils inside sub-plot b2 was labelled with 120 kg N/ha in three split applications of 40 kg N/ha at planting, 40 kg N/ha at 30 days after planting and 40 kg N/ha before silking.

Ordinary Ammonium sulphate was also applied in sub-plots at the same time and the same rate as the labelled fertilizer. The labelled fertilizer was diluted with water and sprinkled onto the soil inside the sub-plots to ensure that the labelled fertilizer penetrated the soil. A one meter perimeter around each sub-plot was also sprinkled with water at the same rate as the sub-plot to ensure moisture equilibration within and outside the sub-plot. Basal application of P and K at the rate of 30 kg P<sub>2</sub>O<sub>5</sub>/ha and 30 kg K<sub>2</sub>O/ha were also applied to all the isotope plots. Two hills of corn (4 plants) were planted in each sub-plot at the same time as the larger (yield) plots were planted.

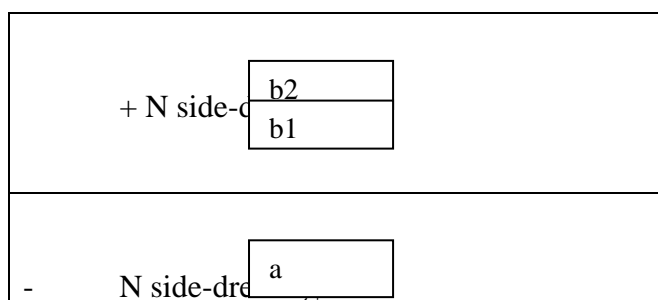


FIG. 2 Micro-plot lay-out for treatments 2, 3 and 6.

In the first fertilizer-manure experiment, corn planting was carried out 2 weeks after manure was incorporated into the plots. Rows were spaced 75 cm apart with 50 cm between seed hills containing two seeds per hill in the rows. Ammonium sulphate (N), superphosphate (P) and muriate of potash (K) was applied at the rate of 20-30-30 kg NPK/ha as basal fertilizers in the designated plots. Side-dressing were applied one month after sowing and at silking at the rate of 40 kg N/ha.application.

All the cultural management and pests control activities such as watering, weeding and spraying of insecticides were done as needed [please describe the practices and the timing along with chemicals and the rates of application]. At cob maturity, whole plants (16 hills) were harvested from each sub-plot for the determination of biomass and grain yield. All whole plants (2 hills/sub-plot) from the  $^{15}\text{N}$ -labeled sub-plots were harvested. The stover (stem, stalk and leaves), corn ears, and roots (excavated from the soil and cleaned) were separated for processing. Fresh weights of each plant constituents were weighed and recorded, then sub-sampled by the quartering, and fresh weights of the sub-sample recorded and oven-dried at  $65^{\circ}\text{C}$  to determine dry matter content.

Soil samples from each sub-plot and sub-plots were collected after harvest and were brought to BSWM Research Laboratory for chemical and physical analyses. Soil samples from each sub-plot were also collected for chemical, physical and  $^{15}\text{N}$  analysis.

## 2.5 Sample Preparation

Plant samples for chemical analyses like total N and  $^{15}\text{N}$  determinations were ground using a Wiley mill. Soil samples were air-dried, pulverized using mortar and pestle and sieved with a 2 mm screen.

### 2.5.1 Total N and $^{15}\text{N}$

Kjeldahl and Dumas dry combustion method were used for total N and  $^{15}\text{N}$  determinations [3].JASCO  $^{15}\text{N}$  emission spectrometer was used to determine  $^{15}\text{N}$  % atom excess.

The data recorded is as follows:

- chemical and physical analyses of the soil before commencing the experiment and after harvest of the first and second (residual) experiments.
- chemical analysis of manure prior to soil application
- $^{15}\text{N}$  analysis of soils in sub-plots
- total N and  $^{15}\text{N}$  analysis of stover, roots, grains using the above mentioned methods and references
- chemical analysis of plant tissue samples
- biomass fresh and dry weight determinations
- total and plant component yields at harvest
- N- uptake of crops from the fertilizer and manure



### 2.5.2 Second experiment (residual fertilizer effects)

In the second experiment, no inorganic or manure fertilizers were applied to estimate the residual effects of manure and inorganic fertilization on the subsequent corn crop. Identical parameters for manure, soil and crop were obtained for both experiments.

## 2.6 Statistical Analysis of Data

The data gathered was analyzed using analysis of variance (ANOVA) procedure for a split-plot, randomized complete block design. The treatment means were compared using a Duncan's Multiple Range Test at the 5% and 1% levels of significance.

## 3. RESULTS

### 3.1 Nutrient compositions of manures applied

Chemical composition of the manures applied as fertilizer in the experiment is shown in Table I. The percentage composition of N, P<sub>2</sub>O<sub>5</sub> and CaO in manures from cattle fed conventional ration and with improved manure management were higher than those other the manure sources.

TABLE I. CHEMICAL ANALYSIS OF THE DIFFERENT MANURES USED IN THE EXPERIMENT

Manure	N (%)	P <sub>2</sub> O <sub>5</sub> (%)	K <sub>2</sub> O (%0)	CaO (%)	MgO (%)	Zn ppm	Cu ppm	Fe ppm	Mn ppm
T3*	1.54	0.82	0.66	0.53	0.54	113	37.5	20750	3325
T4	1.95	1.95	0.35	1.09	4.70	27.5	87.5	7150	4275
T5	1.85	0.98	0.57	0.53	6.01	183	42.5	16900	3025
T6	1.75	1.49	0.38	0.75	7.75	200	60.0	18775	3875

\*where T3 is basal diet with conventional manure storage; T4 is basal diet with improved manure storage; T5 is UMMB with conventional manure storage; and T6 is UMMB with improved manure storage

### 3.2 Effect on the chemical and physical properties of the soil

The chemical and physical property of the soils in the experimental area prior to the application of treatments is shown in Table II.

TABLE II. INITIAL SOIL CHEMICAL AND PHYSICAL CHARACTERISTICS OF THE EXPERIMENTAL AREA

Chemical Characteristics	Physical characteristics
pH (1:1)	Bulk density (g/cc) 1.54
OC (%)	Porosity (%) 41.9
OM (%)	Aggregate stability (mm) 1.04
N (%)	2.11
Olsen P (ppm)	65
Exchangeable bases in meq/100 g soil	
K	0.43
Ca	5.80
Na	1.60
Mg	4.20

TABLE III. MEAN CHEMICAL AND PHYSICAL PROPERTIES OF SOIL BY TREATMENT AND CROPPING

Treatment	Organic Matter		Phosphorus		Potassium		Bulk Density		Porosity		Aggregate Stability	
	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>
	T1	1.26	0.98	26.88	30.01	0.51	0.79	1.53	1.49	40.88	43.90	1.08
T2	1.59	1.10	17.63	26.17	0.45	0.73	1.47	1.55	44.66	43.15	1.02	1.11
T3	1.59	1.10	18.37	32.59	0.48	0.52	1.44	1.53	45.85	42.40	1.18	1.18
T4	1.57	1.20	30.44	31.99	0.39	0.56	1.47	1.60	44.72	39.56	1.05	1.10
T5	1.57	1.34	31.64	26.96	0.49	0.63	1.45	1.59	45.56	40.06	1.08	1.01
T6	1.54	1.35	25.40	28.18	0.43	0.60	1.44	1.54	45.85	41.76	1.05	1.22

### 3.3 Statistical analysis

Variables	F Statistics	Probability
<b><u>Trial 1:</u></b>		
Soil P	1.21	0.332456
Soil K	2.70	0.0439891*
Soil OM	5.63	0.001316*
Bulk Density	1.32	0.289690
Aggregate Stability	0.66	0.658329
Porosity	2.93	0.032218*
Dry Matter Yield	1.98	0.116548
Grain Yield	0.70	0.631543
<b><u>Trial 2:</u></b>		
Soil P	0.53	0.751074
Soil K	2.56	0.053241
Soil OM	2.41	0.064746
Bulk Density	1.84	0.141403
Aggregate Stability	0.77	0.581315
Porosity	2.18	0.088893
Dry Matter Yield	1.30	0.294270
Grain Yield	4.22	0.006400*

\* Term significant at alpha = 0.05

#### Duncan's Multiple-Comparison Test for Trial 1

##### Soil K (meq/100g)

Treatment	Count	Mean	Different From Groups
4	6	0.3866667	1
6	6	0.43	
2	6	0.4533333	
3	6	0.4816667	
5	6	0.4866667	
1	6	0.51	4

There is significant difference among treatments. Treatment 1 is better than treatment 4.

##### Soil OM (Percent)

Treatment	Count	Mean	Different From Groups
1	6	1.263333	6, 4, 5, 2, 3
6	6	1.536667	1
4	6	1.568333	1
5	6	1.576667	1

2	6	1.585	1
3	6	1.591667	1

There is significant difference between treatments. Treatments 2, 3, 4, 5, and 6 are better than treatment 1.

#### Soil Porosity (percent)

Treatment	Count	Mean	Different From Groups
1	6	40.88	2, 4, 5, 6, 3
2	6	44.65333	1
4	6	44.71667	1
5	6	45.59667	1
6	6	45.85	1
3	6	45.85	1

There is significant difference among treatments. Treatments 2, 3, 4, 5, and 6 are better than treatment 1.

#### Grain Yield

Treatment	Count	Mean	Different From Groups
1	6	0.21	2, 4, 6, 5, 3
2	6	0.395	1
4	6	0.3966667	1
6	6	0.4083333	1
5	6	0.425	1
3	6	0.4516667	1

There is significant difference between treatments. Treatments 2, 3, 4, 5, and 6 are better than treatment 1.

In this experiment, the topsoil bulk density (g/cc) declined after the 1<sup>st</sup> trial in Treatments 2-6, but increased during the second cropping (Table III). Statistical analysis showed that the increase in porosity was significantly higher in manured plots over the control. In the second trial, porosity decreased due to the increase in bulk density. It also showed that there were increases in the soil aggregate stability in the manured plots but the increases were not significant over the non-manured plots.

#### 3.3.1 Crop Responses to Manure and N-Fertilizers

The first experiment showed that dry matter yield of plants in the manured-plots and in plots fertilized with inorganic fertilizer was not significantly higher than the non-fertilized control treatment. However, a side dressing of N fertilizer at the rate of 40 kg/ha at 30 days after planting and another 40 kg/ha at silking resulted in higher biomass than corn that did not receive N side-dressing (Table IV). In the second experiment (residual), the same trends were observed in dry matter yield as in the first experiment with non-significant increases in the dry matter yield in treated plots and a significant increase with an N side-dressing.

TABLE IV. MEAN WEIGHT OF DRY MATTER AS AFFECTED BY TREATMENT AND CROPPING FROM EXPERIMENT 1 AND 2 AND THE TOTAL YIELDS FOR THE two EXPERIMENTS

Treatment	First Crop	Second Crop	Combined Yield
T1	4.52±0.61	2.37±0.61	6.88±0.99
T2	5.33±0.74	2.75±0.38	8.08±0.69
T3	5.17±0.42	3.02±0.45	8.18±0.78
T4	4.87±0.35	2.87±0.81	7.73±1.01
T5	5.50±1.32	2.65±0.56	8.15±1.45
T6	5.40±0.57	2.68±0.62	8.08±0.95

No significant increases were obtained in the grain yield of corn with the treatments imposed beyond that achieved by the non-fertilized control during the first experiment. N-side-dressing significantly improved grain yield in corn compared to the section of the plots that did not receive N side dressing. In the second experiment (residual), a significant increase in the grain yield was obtained in T2, T3, T4, T5, and T6 compared to the control (T1). N side-dressed plots from experiment had a significant increase in grain yield compared to the non-side-dressed plants.

TABLE V. MEAN GRAIN YIELD OF CORN FROM EXPERIMENT 1 AND 2 AND THE TOTAL YIELD FOR THE two EXPERIMENTS

Treatment	First Crop	Second Crop	Combined Yield
T1	3.03±0.41	0.21±0.13	3.24±0.17
T2	3.20±0.50	0.40±0.07	3.60±0.59
T3	3.23±0.39	0.45±0.21	3.68±0.55
T4	2.95±0.21	0.40±0.12	3.35±0.27
T5	3.25±0.49	0.49±0.09	3.68±0.54
T6	3.07±0.52	3.07±0.52	3.48±0.56

### 3.3.2 <sup>15</sup>N Data Analysis

### 3.4 Plant N-Uptake. (Isotope plot B2)

The highest plant N-yield was obtained in the treatment with best feeding and manure management practices (T6) although the N-yield was not significantly higher than for the other treatments. Similar trends in yield were observed in the first and second experiments (Table VI).

TABLE VI. MEAN NITROGEN YIELD (KG/HA) OF LABELED CORN PLANT PARTS WHERE T2 IS INORGANIC FERTILIZER; T3 IS MANURE FROM CONVENTIONALLY-FED CATTLE WITH CONVENTIONAL MANURE STORAGE; AND T6 IS SUPPLEMENTED CATTLE WITH IMPROVED MANURE STORAGE

Treatment	Stover	Grain	Roots
<i>First Crop</i>			
T2	22.20±3.22	79.47±17.65	1.88±0.94
T3	21.81±4.13	64.20±18.97	2.78±1.36
T6	26.10±3.81	80.93±30.21	3.94±1.80
<i>Second Crop (Residual)</i>			
T2	6.19±2.74	3.14±4.08	0.42±0.41
T3	3.48±1.57	2.98±0.65	0.41±0.12
T6	8.41±6.29	5.12±2.44	0.76±0.32
<i>Combined Yield</i>			
T2	28.39±5.54	82.61±21.07	2.30±1.34
T3	25.29±5.43	73.38±21.38	3.47±1.36
T6	34.51±7.86	73.09±31.01	4.70±1.66

Using the isotope technique, nitrogen derived from fertilizer (Ndff), fertilizer N Yield (FNY) and Fertilizer Nitrogen Use efficiency were calculated using sub-plots b2. The treatment with improved nutrition and manure management provided fertilizers-N for the first crop with the highest % Ndff (Nitrogen derived from fertilizer) and Fertilizer Nitrogen Yield (FNY) in the grains and stover but was not significantly different to that from inorganic fertilizers N source and conventional manure storage (Tables VII and VIII). The Ndff for the inorganic fertilizer treatment (T2) was slightly higher in the roots compared to manure treatments with conventional and improved manure management in the second crop to measure residual fertilizer effects

TABLE VII. MEAN PERCENTAGE OF NITROGEN DERIVED FROM FERTILIZER AND MANURE

Treatment	Stover	Grain	Roots
<i>First Crop</i>			
T2	4.64±0.86	29.95±1.50	4.55±0.70
T3	4.67±1.02	29.28±1.73	4.87±0.75
T6	4.82±0.48	30.09±0.88	4.07±0.56
<i>Second Crop (Residual)</i>			
T2	1.45±0.72	1.35±1.04	1.60±1.16
T3	1.30±1.01	1.92±1.05	1.89±0.66
T6	1.19±0.76	1.38±0.07	2.34±0.30
<i>Combined Yield</i>			
T2	6.09±0.33	31.30±0.46	6.15±0.57
T3	5.97±1.68	32.04±2.35	6.76±0.78
T6	6.01±0.96	31.91±0.55	6.41±0.81

TABLE VIII. MEAN FERTILIZER NITROGEN YIELD (KG/HA) OF CORN PLANTS PARTS

Treatment	Stover	Grain	Roots
<i>First Crop</i>			
T2	1.05±0.34	23.85±5.83	0.09±0.06
T3	1.04±0.40	18.81±5.65	0.13±0.05
T6	1.27±0.30	24.18±8.46	0.16±0.06
<i>Second Crop (Residual)</i>			
T2	0.09±0.06	0.02±0.02	0.007±0.006
T3	0.06±0.06	0.06±0.04	0.014±0.007
T6	0.13±0.16	0.07±0.04	0.018±0.010
<i>Combined Yield</i>			
T2	1.14±0.33	23.88±5.84	0.93±0.06
T3	1.10±0.43	21.14±5.68	0.14±0.05
T6	1.40±0.36	20.73±8.34	0.18±0.06

Fertilizer Nitrogen Use Efficiency (Table IX) showed a similar pattern to the results obtained for Ndff for the component parts of corn plants. Residual fertilizer effects in the second crop were lower for nitrogen recovery in corn plant parts and whole plants.

TABLE IX. MEAN PERCENTAGE OF FERTILIZER NITROGEN USE EFFICIENCY (% FNUE) OF CORN PLANT

Treatment	Stover	Grain	Roots	Whole
<i>First Crop</i>				
T2	0.87±0.29	19.88±4.85	0.07±0.04	20.82±5.06
T3	0.87±0.33	15.68±4.70	0.11±0.04	16.65±5.03
T6	1.06±0.26	20.15±7.05	0.13±0.05	21.34±6.99
<i>Second Crop (Residual)</i>				
T2	0.08±0.06	0.02±0.03	0.01±0.00	0.11±0.09
T3	0.05±0.05	0.05±0.04	0.01±0.01	0.11±0.10
T6	0.11±0.14	0.06±0.03	0.02±0.01	0.19±0.18

#### Plant N-Uptake (isotope plot A and B1)

Isotope dilution techniques or indirect labelling techniques were used to quantify the amount of nitrogen contributed by the manure, however, insufficient data was available to measure the amount of N contributed by the manure using these techniques which were further elaborated in the discussion of results.

In isotope plots b1 and a where the soil was labelled with 20 kgN/ha <sup>15</sup>N ammonium sulphate using indirect labelling technique. Treatments with manure and no manure and fertilizer N were set up. The control was that of treatment labelled with 20kg<sup>15</sup>N/ha ammonium sulphate and no other source of nitrogen was added.

In sub-plot b1 (with supplemental N) application of manure with improved manure management resulted in the highest N yield in stover of the treatments measured. Results obtained from inorganic fertilizer application and conventional storage were comparable. In corn grain, the treatment receiving organic fertilizers alone had the highest N yields but was not significantly different from the manure treatment with improved manure management or conventional manure storage (Table X). A similar trend was observed for Ndff, FN yield and Fertilizer Nitrogen Use Efficiency in Table XI.

TABLE X. MEAN N YIELD FROM STOVER AND GRAIN IN THE FIRST EXPERIMENT

Treatment	Stover	Grain
T2	15.8	56.9
T3	14.3	32.7
T6	18.5	43.3

TABLE XI. PERCENT NDFD, FNY AND FNUE OF STOVER AND CORN GRAIN IN THE FIRST EXPERIMENT

Treatment	% Ndff		FN yield (kg/ha)		% FNUE	
	Stover	Grain	Stover	Grain	Stover	Grain
T2	30.3	27.3	4.96	14.4	4.13	
T3	15.2	27.6	3.26	9.2	2.16	
T6	17.2	28.4	3.94	13.0	4.78	

#### 4 DISCUSSION

The application of manures or organic materials to soil to improve the physical properties of soils is a well-established practise in crop production. The fibrous materials in manure have been shown to play a key role in altering the physical properties of soils. In these experiments, the topsoil bulk density declined after the first cropping in the fertilizer treatments (T2-T6), but increased again during the second crop (Table III). The decrease in bulk density maybe attributed to the direct effect of manure application brought about by an increase in the volume of micropores [this does not account of the effect in T2 of inorganic fertilizers]. The increase in bulk density during the second trial (residual) show that the fertilizer applications applied may not be enough to ensure a sustained change in soil properties with ongoing corn cropping. With the decrease of bulk density in fertilized soils after the first crop there was a corresponding increase in soil porosity (Table III). This was also observed in green manure as a source of organic fertilizer, reduces soil bulk density as it enhanced total soil porosity [4].

There were increases in the soil aggregate stability in the manured plots as shown in Table III, but the increases were not significantly greater than for the non-manured plots. This maybe attributed to the formation of humic substances that are relatively stable binding agents in soils ([5, 6].

Due to severe drought and water shortage, the quantification of nitrogen uptake by plants from inorganic fertilizers and manures was not possible in these experiments. The use of  $^{15}\text{N}$  labelling techniques provided significant information on fertilizer use efficiency and indirect isotope dilution techniques provided important information on animal manure application and storage. Fertilizer and manuring experiments have indicated that the environment loss of N is strongly influenced by soil factors, weather and the amount, timing and methods of application [7].

Prasatsrisupab et al., failed in a study undertaken to calculation the NdfSS for nitrogen derived from sewage sludge. The results of their experiments showed that the nitrogen derived from sewage sludge can only be determined by applying a higher rate of fertilizer. NdfSS in maize was calculated using the N-balance. Another reason that attributed to the results of the experiments was probably a very short equilibrium made during the pre-labeling of soil and the low amount of ammonium sulphate used. Application of  $^{15}\text{N}$  labelled fertilizer and residues at the same time has been shown to cause errors associated with pool substitution [9, 10]. One possible explanation might be that the microbial biomass became a large sink for applied N and that through pool substitution, unlabelled N was substituted for  $^{15}\text{N}$  labelled N and the unlabelled N became available for crop uptake [11]. These problems can be overcome if the soil is pre-labelled with  $^{15}\text{N}$  and left to equilibrate for up to 6 months prior to application of residues [10].

No fertilizers were applied to any of the treatments for the second crop to determine the residual effects of the original fertilizer treatments. There was a marginal carry over of fertilizer effects for the second crop resulting in poor yields across all fertilizer treatments but these yields were [significantly] higher than the original non-fertilized treatment. The corn plants were observed during the growing period to be stunted and exhibited a yellowing of the leaves. This typically meant that the crop has a deficiency in nitrogen [12]. This is consistent with poor plant nutrition leading to reduced plant and grain.

## 5 CONCLUSION

Animal manure is an important natural resource in a sustainable livestock-crop farming system. Integrating manure, as a fertilizer, into crop production is considered the primary mechanism for manure management and disposal. However, to successfully utilize manure as a fertilizer will require assessing the available nutrients in the manure, and calculating the appropriate application rates to ensure that the nutrient needed for the crop are met. It is concluded from these studies that further work is required on: (i) mineralization of nutrients in manure, (ii) using  $^{15}\text{N}$  labelling to determine fate of N from manures in the system and (ii) manure application rates.

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