

## Effect of Mulberry leaf pellet (MUP) supplementation on rumen fermentation and nutrient digestibility in beef cattle

Nguyen Thi Huyen<sup>1</sup> Metha Wanapat<sup>1</sup>

<sup>1</sup>Tropical Feed Resource Research and Development Center (TROFREC), Department of Animal Science, Faculty of Agriculture, Khon Kean University, Thailand  
123 Mittaparb Rd., Maung District, Khon Kaen 40002, Thailand  
Telephone/Fax : (+66)-043-20 2368  
E-mail: metha@kku.ac.th

### Abstract

This study was designed to determine the effect of mulberry leaf pellets (MUP) on feed intake, digestibility and rumen fermentation in beef cattle. Four, ruminally fistulated crossbred (Brahman x Thai native) beef cattle with initial body weight (BW) of 420±15 kg were randomly assigned according to a 4 x 4 Latin square design. The dietary treatments were different levels of MUP supplementation and were as follows; T1 = 0 g MUP/head/day; T2 = 200 g MUP/head/day; T3 = 400 g MUP/head/day; T4 = 600 g MUP/head/day. All animals were kept in individual pens and concentrates were offered at 0.5% BW/day. The experiment was conducted for 4 periods, and each period lasted for 21 days. During the first 14 days, all animals were fed with respective diets and rice straw was fed *ad libitum*. During the last 7day, the animals were moved to metabolism crates for total urine and fecal collection. Roughage and total dry matter intakes significantly increased ( $P<0.05$ ) when MUP were supplemented, and was highest with supplementation at 600 g MUP/head/day. Apparent digestibilities of dry matter, organic matter, crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF) were improved ( $P< 0.05$ ) when increasing level of MUP, especially T4. Ruminal pH and temperature were similar among treatments ( $P>0.05$ ). Propionate (C3) and butyrate (C4) proportions tended to reduced while total VFA, acetate and C2:C3 ratio increased when increasing level of MUP supplementation. Therefore, MUP can be used as a protein source for ruminants and supplementation at 600 g/head/d could improve rumen fermentation efficiency and digestibility.

**Keywords:** Mulberry leaf pellet, rice straw, digestibility, rumen fermentation

### 1. Introduction

Low-quality roughages in the tropics are generally low in nitrogen. Supplementation with protein sources has been shown to increase the utilization of low quality roughages in ruminants [1]. However, protein-rich feed prices such as soybean meal is increasing. Thus, scientists have been studying

and looking for easy, cheap and sustainable protein sources in tropical area. Urea (NPN) is cheaper than soybean meal and it has been using to improve low quality of roughages by mixing in concentrate [2]. Mulberry (*Morus alba*) can grow well under varied climatic conditions, ranging from temperate to tropical. Mulberry leaves have a high crude protein content (18 to 25%) [3], high palatability and high intake. Moreover, mulberry has 50% of rumen undegradable protein and net energy for milk production (1.48 Mcal/kg) [4]. High quality feed pellets have shown to increase feed intake, rumen fermentation end – products as well as increasing milk yield [5]. Moreover, dry matter intake, digestibility of nutrients, volatile fatty acids were improved when using mulberry leaf pellets with 3% urea as a protein source replacement for soybean meal in concentrate [6]. However, using mulberry leaf pellets with urea in cattle have been rather limited.

### 2. Objective

This experiment was conducted to investigate the effect of mulberry leaf pellet (MUP) supplementation on rumen fermentation and nutrient digestibility in beef cattle fed rice straw.

### 3. Materials and methods

The experiment was conducted at Tropical Feed Resource Research and Development Center (TROFREC), Department Animal Science, Faculty of Agriculture, Khon Kean University, Thailand from April to August, 2010.

**Table 1.** Ingredients and chemical compositions of diets used in the experiment.

Item	Ratio		
	Conc. <sup>2</sup>	MUP <sup>1</sup>	Rice straw
<b>Ingredients</b>			
Cassava chip	75.0	-	
Mulberry meal	-	82.0	
Cassava starch	-	0.5	
Rice bran	6.0	-	
Coconut meal	5.0	-	
Palm meal	6.5	-	
Urea	3.5	10.0	
Molasses	1.0	4.5	
Sulfur	1.0	1.0	
Mineral premixed	1.0	1.0	
Salt	1.0	1.0	
<b>Chemical composition, %</b>			
DM	94.1	92.3	96.0
-----%DM-----			
OM	92.5	88.2	86.2
Ash	7.5	11.8	13.8
CP	14.2	48.7	3.9
NDF	17.4	20.4	75.9
ADF	11.5	14.5	47.3
TDN	70.2	69.2	50.1

<sup>1</sup> MUP: Mulberry pellets with 10% urea; <sup>2</sup> Conc.: Concentrate, DM: dry matter, OM: organic matter, CP: crude protein; NDF: neutral detergent fiber; ADF: acid detergent fiber; TDN: Total digestible nutrients.

Four, ruminally fistulated crossbred (Brahman x Thai native) beef cattle with initial BW of 420 ± 15kg were randomly assigned to receive four dietary treatments according to a 4 x 4 Latin square design. The feed ingredients and chemical composition of experimental diets, MUP, and rice straw are shown in Table 1. All animals were kept in individual pen, mineral block, fresh water and rice straw were available for ad libitum consumption. Concentrate (14.2% CP) was fed for animals with 0.5% of BW/day. The four treatments were as follows: T1 = 0 g MUP/head/day; T2 = 200 g MUP/head/day; T3 = 400 g MUP/head/day; T4 = 600 g MUP/head/day. The MUP was produced according to Wanapat et al. [5]. In brief, (1) collecting mulberry leaves and sun dried about 3 days; (2) mulberry leaves were ground (1mm screen using Cyclotech Mill, Tecator, Sweden); (3) mixed mulberry leaf meal with urea, cassava starch, molasses, salt, mixed mineral and sulfur in respective ratio (Table 1); (4) then mixed well all ingredients in step (3) added water with ratio of 0.8:1 (water and mixture, respectively); (5) made pellets using machine and then sun dried 3 days. The experiment was conducted for 4 periods and each period lasted 21days. During the first 14 days, all animals were fed with respective diets whereas during the last 7 days, the animals were moved to metabolism crates for total collection of urine and feces.

Feeds were sampled and fecal samples were collected from the total collection of each individual cattle on each treatment during the last 7 days of each period at morning and afternoon feeding. Feed and fecal were analyzed for DM, ash, CP content [7], NDF and ADF [8]. At the end of each period, rumen fluid was sampled at 0, 2, 4 and 6 h after feeding. Temperature and pH of rumen fluid were measured using a portable pH and temperature meter immediately after rumen fluid was collected. Rumen fluid samples were used for NH<sub>3</sub>-N analysis using the micro-Kjeldahl methods [9] and VFA analysis using HPLC [8]. All data were analyzed in a 4 x 4 Latin Square design by analysis of variance run in the GLM procedure of Statistical Analysis [10]. Treatment means were statistically compared by Duncan's New Multiple Range Tests.

#### 4. Results

Chemical compositions of concentrate, MUP10 and rice straw are shown in Table 1. Concentrate, pellets and rice straw contained 14.2%, 48.7% and 3.9% of CP, respectively. The effects of MUP supplementation on total dry matter intake and nutrient digestibility are presented in Table 2. It was found that roughage and total dry matter intakes in terms of kg/d and %BW were increased (P<0.05) with increasing level MUP supplementation.

**Table 2.** Effect of MUP supplementation on feed intakes and nutrient digestibility in beef cattle.

Item	Supplementation of MUP <sup>1</sup> (g/head/d)				SEM
	0	200	400	600	
<b>Rice straw DM intake</b>					
kg/day	6.6 <sup>a</sup>	6.8 <sup>b</sup>	7.0 <sup>b</sup>	7.3 <sup>c</sup>	0.05
%BW	1.6 <sup>a</sup>	1.6 <sup>a</sup>	1.7 <sup>b</sup>	1.7 <sup>b</sup>	0.03
<b>Concentrates DM intake</b>					
kg/day	2.0	2.0	2.0	2.0	0.02
%BW	0.5	0.5	0.5	0.5	0.003
<b>Total DM intake</b>					
kg/day	8.6 <sup>a</sup>	9.0 <sup>b</sup>	9.4 <sup>c</sup>	9.9 <sup>d</sup>	0.05
%BW	2.0 <sup>a</sup>	2.1 <sup>a</sup>	2.3 <sup>b</sup>	2.3 <sup>b</sup>	0.04
<b>Average daily gain</b>					
kg/day	0.1 <sup>a</sup>	0.3 <sup>a</sup>	0.4 <sup>ab</sup>	0.8 <sup>b</sup>	0.14
<b>Apparent digestibility, %</b>					
DM	66.3 <sup>a</sup>	66.8 <sup>a</sup>	69.5 <sup>b</sup>	69.8 <sup>b</sup>	0.54
OM	69.4 <sup>a</sup>	69.6 <sup>a</sup>	72.4 <sup>b</sup>	72.9 <sup>b</sup>	0.49
CP	54.1 <sup>a</sup>	61.0 <sup>b</sup>	67.9 <sup>c</sup>	69.4 <sup>c</sup>	0.80
NDF	58.3 <sup>a</sup>	59.5 <sup>b</sup>	63.8 <sup>c</sup>	66.0 <sup>d</sup>	0.19
ADF	55.1 <sup>a</sup>	56.3 <sup>ab</sup>	58.3 <sup>ab</sup>	59.9 <sup>c</sup>	0.60

<sup>a, b, c, d</sup> Means in the same row with different superscripts differ (P<0.05), <sup>1</sup> MUP: Mulberry pellets with 10% urea; DM: dry matter; OM: organic matter; CP: crude protein; NDF: neutral detergent fiber; ADF: acid detergent fiber.

Apparent digestibility (%) of DM, OM, CP, NDF and ADF were highest when supplementation of MUP at 600 g/head/d ( $P<0.05$ ). Effect of MUP supplementation on rumen fermentation characteristic in cattle is shown in Table 3. Ruminal pH and temperature were not altered among treatments and the values were stable at pH 6.3 and temperature 38.6 to 38.9°C. Ruminal  $\text{NH}_3\text{-N}$  concentration were significantly different among treatments ( $P<0.05$ ). There were significant differences in acetic acid, propionic acid and butyric acid proportions and acetic: propionic ratio ( $P<0.05$ ) (Table 3). Moreover, total VFA concentration increased ( $P<0.05$ ) from 98.2 to 104.3 mM/l when increasing level of MUP from 0 to 600 g/head/day, respectively.

**Table 3.** Effect of MUP supplementation on rumen fermentation characteristics in beef cattle.

Item	Supplementation of MUP <sup>1</sup> (g/head/d)				SEM
	0	200	400	600	
Rumen parameters					
pH	6.3	6.3	6.3	6.3	0.03
Temperature, °C	38.8	38.6	38.9	38.8	0.08
$\text{NH}_3\text{-N}$ , mg/dl	10.7 <sup>a</sup>	13.6 <sup>ba</sup>	16.6 <sup>cb</sup>	18.5 <sup>c</sup>	0.99
TVFA, mMl	98.2 <sup>a</sup>	101.2 <sup>b</sup>	102.8 <sup>c</sup>	104.3 <sup>d</sup>	0.18
C2%	67.4 <sup>a</sup>	68.6 <sup>b</sup>	68.5 <sup>b</sup>	69.2 <sup>c</sup>	0.14
C3%	21.2 <sup>c</sup>	20.4 <sup>b</sup>	20.3 <sup>ab</sup>	20.0 <sup>a</sup>	0.11
C4%	11.4 <sup>c</sup>	11.0 <sup>ab</sup>	11.2 <sup>ab</sup>	10.8 <sup>a</sup>	0.09
C2:C3	3.2 <sup>a</sup>	3.4 <sup>b</sup>	3.4 <sup>bc</sup>	3.5 <sup>c</sup>	0.02

<sup>a, b, c</sup> Means in the same row with different superscripts differ ( $P<0.05$ ), <sup>1</sup> MUP: Mulberry pellets with 10% urea; TVFA: Total volatile fatty acids;  $\text{NH}_3\text{-N}$ : ammonia nitrogen; C2: acetate; C3: propionate; C4: butyrate; C2:C3: acetate: propionate ratio.

## 5. Discussions

Roughage, total dry matter intake and nutrient digestibility were increased when increasing MUP supplementation at 200, 400, 600 g/head/day and were agreed with report by Bodthaisong [6]. Ruminal pH and temperature were within the range considered for optimal microbial digestion of fiber and protein (6.0-7.0; [12], [1]). Ruminal  $\text{NH}_3\text{-N}$  concentrations were 10.7 - 18.5 mg/dL and were in optimal ruminal  $\text{NH}_3\text{-N}$  ranges (15- 30mg/dl, [2]) for improving rumen ecology, microbial protein synthesis, digestibility and voluntary feed intake. The observed increasing in acetate proportion was associated with increasing roughage intake. The results of total VFA concentration in all diets were found in normal concentrations (70 – 130 mM/l, [13]).

## 6. Conclusions

Based on this experiment, it could be concluded that MUP is potential to be used as a protein sources for ruminants. Feeds intake, rumen fermentation and nutrient digestibility were significantly improved by supplementation of MUP. Therefore, MUP is recommended for supplementation at 600 g/head/d for cattle fed on low-quality roughages such as rice straw.

## 7. Acknowledgements

The authors would like to express their most sincere thanks to the Tropical Feed Resources Research and Development Center (TROFREC), Department of Animal Science, Faculty of Agriculture, Khon Kaen University, Thailand and TRIG project for their kind financial support and the use of the research facilities.

## 8. References

- [1] Wanapat, M., 1990. Nutrition Aspect of ruminant Production in southeast Asia with Special Reference to Thailand. Funny Press, Ltd., Bangkok, Thailand, ISBN: 9746766198, pp: 125- 136.
- [2] Wanapat, M. and O. Pimpa, 1999. Effect of ruminal  $\text{NH}_3\text{-N}$  levels on rumen fermentation, purine derivatives, digestibility and rice straw intake in swamp buffaloes. Asian-Aust. J. Anim. Sci., 12: 904-907.
- [3] Ba, N. X., V. D Giang, and L. D Ngoan., 2005. Ensiling of mulberry foliage (Morus alba) and the nutritive value of mulberry foliage silage for goats in central Vietnam. Live stock Research for Rural Development. 17(15) .[http://www.cipav.org.co/lrrd/lrrd17/ba\\_17015.htm](http://www.cipav.org.co/lrrd/lrrd17/ba_17015.htm)
- [4] Makkar, H.P.S., B. Singh and S.S. Negi, 1989. Relationship of rumen degradability with microbial colonization, cell wall constituents and tannin levels in some tree leaves. Anim. Prod. Sci., 49: 299-303.
- [5] Wanapat, M., K. Sommart., O. Pimpa, and S. Boonsorn., 1996. Supplementation of high quality feed pellet to increase milk productivity at small-holder farmers level. In: Proceeding of the 8<sup>th</sup> AAAP Animal Science Congress. October 13-18, 1996. Japanese Society of Zootechnical Science, Tokyo, Japan.
- [6] Bodthaisong, S. 2008. Effect of Mulberry leaf pellet as a protein source in replacement for soybean meal in concentrate for lactating dairy cows. Master of Science. Thesis in Animal Science, Graduate school, Khon Kaen University, Thailand, pp: 41-46.

- [7] AOAC., 1997. Official Methods of Analysis. 16<sup>th</sup> ed. Association of Official Analytical Chemists. Gaithersburg, MD.
- [8] Van Soest, P. J., J. B. Robertson, and B. A. Lewis., 1991. Method for dietary fiber, neutral detergent fiber and non-starch polysaccharide in relation to animal nutrition. *J. Dairy Sci.* 74, 3583-3597
- [9] AOAC., 1995. Official Methods of Analysis, Animal Feeds. 16<sup>th</sup> Edn, Association of Official Analytical Chemists, Virginia, USA., pp: 1-18.
- [10] Samuel, M., S. Sagathewan, T. Thomas and G. Mathen, 1997. An HPLC method for estimation of volatile fatty acids of ruminal fluid. *Indian J. Anim. Sci.*, 69: 805-807.
- [11] SAS., 1998. SAS/STAT User's Guide. Version 6.12. SAS Inst. Inc., Cary, NC.
- [12] Lyle, R.R., R.R. Johnson, J.V. Wilhite and W.R. Backus, 1981. Ruminant characteristics in steers as affected by adaptation from forage to all concentration diets. *J. Anim. Sci.*, 53: 1383-1394.
- [13] France, J. and R.C. Siddons., 1993. Volatile fatty acids production. In: *Quantitative Aspects Ruminant Digestion and Metabolism* (Eds., J.M. Forbes and J. France). C. A. B. International, Willingford, UK.