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Effect of Cassoy-urea Pellet as a Protein Source in Concentrate on Ruminal Fementation and Digestibility in Cattle

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ABSTRACT : Four male crossbred native beef cattle (average body weight of 427.7 kg) were randomly allocated to four types of cassoy-urea pellet as a source of protein in concentrate according to a 4×4 Latin square design to determine effect of diets on ruminal fermentation and nutrient digestibility. The four types of cassoy-urea pellets contained cassava hay, soybean meal, urea and binding agent at 79.2:19.8:0:1 (27.9% CP dry matter), 78.4:19.6:1:1 (30.4% CP), 77.6:19.4:2:1 (33.0% CP) and 99:0:0:1 (23.8% CP) for dietary treatments; 1, 2, 3 and 4, respectively. All four concentrate mixtures contained similar crude protein levels (11% CP) and were fed to animals in two equal parts (0.5% of body weight per day) while urea-treated rice straw (5% urea) was given *ad libitum*. The experiment revealed that dietary concentrate treatments had no effect on dry matter intake while digestibilities of neutral-detergent fiber and crude protein were higher ($p<0.05$) in cattle fed dietary treatments 1, 2 and 3 than in cattle fed dietary treatment 4. Ruminal ammonia-nitrogen (NH_3-N), was higher and acetic acid concentration (C2) and ratio of C2 to propionic acid (C3) were lower ($p<0.05$) in cattle fed dietary treatments 1, 2 and 3 than in those on treatment 4. It is concluded that use of cassoy-urea pellet as a protein source in concentrates for cattle resulted in improvement of digestibility, ruminal fermentation and rumen ecology. Further research using cassoy-urea pellet in feeding trials with milking cows and fattening beef should be undertaken. (**Key Words :** Cassava Hay, Cassoy-urea Pellet, Rumen Fermentation, Volatile Fatty Acids, Urea-treated Rice Straw, Cattle, Rumen Ecology)

INTRODUCTION

Cassava *(Manihot esculenta, Crantz*) is grown widely in tropical countries. This plant is well known for its adaptability to poor soil condition, drought resistance and pest tolerance. Usually, cassava is grown for root production and is regarded as a cash crop. In addition, supplementation of concentrate containing a high levels of cassava chip with malate could improve rumen fermentation efficiency and rumen microbial protein synthesis in dairy cows (Khampa et al., 2006, 2006a). However, attention has recently been focused on the potential use of the whole cassava crop as cassava hay (CH) in livestock production (Wanapat et al., 1997; 2000a, b). It has been found that cassava hay contained up to 25% crude protein (CP) with a good profile of amino acids (Wanapat et al., 1997; Wanapat, 2003). The use of CH in dairy cattle feeding has been successfully implemented in several ways (Wanapat et al., 2000ab; Wanapat, 2003), such as inclusion in the concentrate supplement (Bezkorowajnyi et al., 1986; Wanapat et al., 1992) or in a high-quality feed block (Koakhunthod et al., 2001). Cassava hay contains condensed tannins (CT) or proanthocyanidin which are common in tropical plants. Condensed tannins are polyphenolics which can easily be solubilized in water and can precipitate protein. The presence of condensed tannins and protein could form a tannin-protein complex by hydrogen-bonding especially under alkaline pH conditions. Tannin-protein complex will be maintained at pH 3.5-7 and will dissociate at pH<3.0 and >0.8 (Jones and Mangan, 1977). Barry and Manley (1984) and Reed (1995) reported that if condensed tannins in the feed exceeded 6% of dry matter, feed intake and digestibility would be dramatrically reduced. If the condensed tannin level was between 2-4% dry matter it would help to protect protein from rumen digestion, thereby increasing by-pass protein or rumen undegradable protein (RUP). Since cassava hay contains 3-4% of condensed tannins, it could form tannin-protein complexes which could act as a source of RUP (Wanapat,

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Table 1. Ingredients and crude protein (CP) content of cassava hay pellet (CHP) and cassoy (cassava+soybean meal) pellet containing urea (cassoy-urea pellet) at 0 (CSU0), 1 (CSU1) and 2% (CSU2)

Type	Ingredient (% dry matter)				% CP
	Cassava hay	Soybean meal	Urea	Binding agent	
CSU0 (T1)	79.2	19.8	0.0	1.0	27.9
CSU1 (T2)	78.4	19.6	1.0	1.0	30.4
CSU2 (T3)	77.6	19.4	2.0	1.0	33.0
CHP (T4)	99.0	0.0	0.0	1.0	23.8

2003). As reported by Wanapat et al. (1997) and Promkot and Wanapat (2004) the rumen by-pass protein of cassava hay was 45.4% of CP content when studied *in situ* in dairy cattle fed urea-treated rice straw.. The most significant improvement from cassava hay supplementation (up to 2 kg/hd/d) was to reduce concentrate use by 42%, which resulted in higher income for small-holder dairy farmers (Wanapat, 2004). However, major objectives in protein nutrition of ruminants not only concern rumen by-pass protein but aim to optimize rumen degradable protein (RDP) for maximizing ruminal formation of high quality microbial protein as well. Ruminants in the tropics normally are fed on low-quality roughages, especially agricultural crop-residues such as rice straw (Doyle et al., 1986; Devendra, 1992), while critical rumen NH_3-N levels for microbial activities were found at 5-20 mg/dl (Perdok and Leng, 1989). Broderick (2005) stressed that urea supplementation appeared to be most effective for improving intake and digestibility of straws and other low quality forages when ruminal ammonia concentrations were very low. Urea N is converted via ammonia into microbial protein thereby supplying additional protein to the host animals. However, intermittent feeding of urea normally leads to high peaks of NH_3-N shortly after ingestion and much lower concentration afterwards. Strategies to feed rumen bacteria have included continuous supply of NPN to keep a high level of ammonia N in the rumen, and introducing to the diet key amino acids, degradable carbohydrates, and mineral salts. Since urea is cheap it could be used for tropical ruminant production, providing it is controlled to enable slow release and/or synchronized with soluble carbohydrate in the rumen, and should be of great value in improving N utilization. Adding controlled-release urea to a high fiber, low crude protein diet in sheep increased NDF digestion and microbial protein formation in the rumen (Puga et al., 2001). Making a combination of cassava hay and urea as a pellet might be a suitable method to control urea release and provide a good source of RDP and RUP. However, a number of *in vitro* studies have also shown improved microbial growth with amino acid and peptide supplementation (Broderick, 2005). In addition, Oltjen (1969) summarized a number of trials showing that when all of the dietary CP was supplied by urea, growth rate, feed efficiency and N-retention in ruminants were about 65% of their values when the same diets contained CP from only isolated soy protein. Therefore cassoy-urea pellets (the pellet mixture of cassava hay, soybean meal, urea and some binding agent) were formulated to contain both RDP and RUP at various levels and were investigated in concentrate mixtures for cattle.

MATERIALS AND METHODS

Animals and design

Four, male fistulated crossbred Brahman-Thai native beef cattle (body weight 427.7±10 kg) were randomly allocated to four types of cassoy-urea pellet as sources of protein in concentrate according to a 4×4 Latin square design to determine effect of diets on ruminal fermentation and nutrient digestibility. Experimental periods were 21 d in length for each period. The cattle were individually penned and clean fresh water and mineral blocks were offered free choice.

Experimental feeds and management

The four types of cassoy-urea pellets as protein sources in concentrate mixtures containing CH, SBM and urea are shown in Table 1.

Ingredient compositions of concentrate feed, CH, SBM and roughage (urea-treated rice straw 5% urea; UTRS) are shown in Table 2. All concentrate mixtures had similar CP by using four types of cassoy-urea pellet as protein sources and were given to animals in two equal parts (0.5% of body weight per day) while urea-treated rice straw (5% urea) was given *ad libitum.*

Sampling procedure, data collection and chemical composition analysis

Urea-treated rice straw, CH, SBM and concentrate were sampled daily during the collection period and were composited by period prior to analyses. Feed and fecal samples were collected during the last five days of each period. Fecal samples were collected by rectal sampling during the last 2 days of sampling. Composited samples were dried at 60°C and ground (1 mm screen using Cyclotech Mill, Tecator, Sweden) and then analysed for DM, ash and CP content (AOAC, 1990), NDF and ADF (Goering and Van Soest, 1970) and acid-insoluble ash (AIA) (Van Keulen and Young, 1977).

Table 2. Ingredient mixtures (%) and chemical compositions of experimental concentrates, urea-treated rice straw (UTRS), soybean meal (SBM) and cassava hay (CH)

Item	CSU0	CSU1	CSU2	CHP	UTRS	SBM	CH
Cassava chip	49.5	51.5	53.5	45.6			
Fine rice bran	14.3	15.4	16.4	9.2			
CSU0	31.1	-	-	-			
CSU1	-	28.1	-	-			
CSU2	-	-	25				
CHP		-	-	40.1			
Molasses	1.7	1.7	1.7	1.7			
Tallow	2.2	2.2	2.2	2.2			
Sulphur	0.6	0.6	0.6	0.6			
Mineral mix	0.6	0.6	0.6	0.6			
% Dry matter							
Dry matter (%)	86.9	86.4	87.0	88.3	51.6	92.6	92.5
Organic matter (%)	90.8	92.1	91.1	91.6	86.5	93.5	92.8
Crude protein (%)	10.9	10.5	10.6	10.7	8.3	44.1	24.5
Neutral-detergent fiber (%)	24.1	20.2	22.2	28.6	79.7	12.3	57.2
Acid-detergent fiber (%)	14.1	11.8	13.9	18.8	53.3	8.4	34.3
Ash (%)	8.2	7.8	8.3	8.4	13.5	6.5	7.2

CSU0, CSU1 and CSU2 = Cassoy contained urea 0, 1 and 2% respectively; CHP = Cassava hay pellet.

The AIA was used to estimate digestibility of nutrients (Van Keulen and Young, 1977). Rumen fluid and jugular blood samples were collected at 0 and 4 h-post feeding. Approximately 200 ml of rumen fluid was taken from the middle part of the rumen at each time at the end of each collection period. Rumen fluid was immediately measured for pH and temperature using a portable pH meter. Rumen fluid samples were then filtered through four layers of cheesecloth. Samples were used for NH_3-N and ruminal volatile fatty acids (VFAs) analyses where 5 ml of H_2SO_4 solution (1M) was added to 50 ml of rumen fluid. The mixture was centrifuged at 16,000×g for 15 minutes and the supernatant stored at -20°C prior to NH_3-N analysis using the micro-kjeldahl method (AOAC, 1990) and VFA analyses using HPLC (Samuel et al., 1997).

Samples of jugular blood were drawn into serum separation tubes at the time of rumen fluid sampling and centrifuged for 20 min at 850×g. The supernatant was decanted and frozen (-20°C) until it was analyzed for blood urea nitrogen (BUN) according to the method of Crocker (1967).

Statistical analysis

Statistical analyses were performed using the GLM procedure of SAS (1998). Data were analyzed using the model $Y_{ijk} = \mu+M_i+A_j+P_k+\varepsilon_{ijk}$. where Y_{ijk} is the observation from animal j, receiving diet i, in period k; μ, the overall of mean, M_i, the mean effect of level of concentrate (i = 1, 2, 3, 4), A_j, the effect of animal (j = 1, 2, 3, 4), P_k, the effect of period (k = 1, 2, 3, 4), ε_{ijk} the residual effect. Mean differences with a significant F value ($p<0.05$) for treatment were statistically compared using Duncan's New Multiple Range Test (Steel and Torrie, 1980).

RESULTS AND DISCUSSIONS

Effect on rumen fermentation and blood metabolites

Data on intakes, ruminal fermentation parameters and blood urea nitrogen are presented in Table 3. For both concentrate and UTRS intakes, there were no significant differences among treatments.

Ruminal NH_3-N and BUN were similar in cattle fed concentrate containing CSU0, CSU1 and CSU2 and were significantly higher than values in the CHP group. Ruminal pH was similar among treatments and was in agreement with a previous study (Wanapat et al., 2004) which revealed that increasing CH:SBM ratio in concentrate reduced concentrations of ruminal NH_3-N and BUN. It was reported that rumen NH_3-N concentrations increased linearly with increasing supplemental RDP levels (Nolte et al., 2003). It was also reported by Wanapat et al. (1997) that cattle fed on cassava hay had lowered rumen NH_3-N and BUN concentration, demonstrating the effect of condensed tannins in forming tannin-protein complex.

Acetic acid concentration (C2) and ratio of C2 to propionic acid (C3) were lower ($p<0.05$) in cattle fed dietary treatments 1, 2 and 3 than that in cattle fed dietary treatment 4 which contained CSU0, CSU1, CSU2 and CHP, respectively. This could be due to the higher level of fiber (NDF and ADF) in treatment 4 (CHP).

Effect on feed intake and digestibility of nutrients

Digestibilities of neutral-detergent fiber and CP (Table 4) were higher ($p<0.05$) in cattle fed dietary treatments 1, 2 and 3 than in treatment 4. The reason might be due to level of NH_3-N concentration in the rumen and an optimized rumen ecology. Perdok and Leng (1989) showed that higher levels of rumen NH_3-N (15 to 30 mg %) improved

Table 3. Effect of using of different type of pellet as a protein source in concentrate on dry matter intake, ruminal fermentation, blood-urea-nitrogen (BUN) in beef cattle steers

Item	CSU0	CSU1	CSU2	CHP	SEM
Dry matter intake					
Concentrate					
kg/hd/d	1.9	1.9	1.9	1.9	0.0
% body weight	0.5	0.5	0.5	0.5	0.0
Urea-treated rice straw					
kg/hd/d	9.9	9.0	9.3	9.5	0.3
% body weight	2.3	2.1	2.2	2.3	0.1
Rumen ecology					
pH					
0 h, post-feeding	6.4	6.4	6.5	6.5	0.04
4	6.5	6.5	6.7	6.7	0.06
Mean	6.5	6.5	6.6	6.6	0.05
NH3-N (mg %)					
0 h, post-feeding	8.4	8.4	8.7	7.9	0.4
4	11.0[a]	11.3[a]	11.7[a]	10.0[b]	0.2
Mean	12.9[a]	13.1[a]	13.6[a]	9.0[b]	0.3
Total VFA (mM/ml)	115.0	115.3	117.7	114.9	0.8
VFA (mol/100 mol)					
Acetate (C2)	70.2[a]	70.5[a]	71.0[a]	74.5[b]	0.7
Propionate (C3)	19.1	18.5	18.0	17.1	0.7
Butyrate (C4)	10.7	11.0	11.1	8.7	0.7
C2:C3	3.6[a]	3.8[a]	3.9[a]	4.4[b]	0.4
BUN (mg %)					
0 h, post-feeding	12.0	12.2	12.9	11.0	0.7
4	13.1[ab]	13.0[ab]	14.7[a]	12.4[b]	0.5
Mean	12.6	12.6	13.8	11.7	0.7

[a, b] Means with different superscripts within rows differ ($p<0.05$).
SEM = Standard error of the means, VFA = Volatile fatty acid.

Table 4. Effect of different type of pellet as a protein source in concentrate on nutrient digestion coefficients and digestible intakes

Item	CSU	CSU1	CSU2	CHP	SEM
Digestion coefficients (%)					
Dry matter	64.9	65.1	65.1	65.0	0.1
Organic matter	72.4	72.6	70.0	72.5	0.8
Crude protein	65.8[ab]	64.5[ab]	67.5[a]	62.3[b]	0.8
Neutral-detergent fiber	74.3[a]	77.4[a]	76.0[a]	70.3[b]	0.9
Acid-detergent fiber	67.4	67.6	66.0	67.5	0.5
Digestible intake (kg/hd/d)					
Dry matter	8.0	7.1	7.3	7.4	0.3
Organic matter	7.4	6.9	6.8	7.2	0.2
Crude protein	0.7	0.6	0.7	0.6	0.04
Neutral-detergent fiber	6.2	5.9	6.0	5.7	0.1
Acid-detergent fiber	3.7	3.4	3.4	3.7	0.1

[a, b] Means with different superscripts within rows differ ($p<0.05$).
SEM = Standard error of the means.

digestibility and subsequent feed intakes. Moreover, higher bacterial counts (cellulolytic, proteolytic and amylolytic bacteria) were found (Wanapat, 2003) in cows fed UTRS which had higher levels of ruminal NH_3-N than cows fed untreated-rice straw.

CONCLUSIONS AND RECOMMENDATIONS

Based on this experiment, the results suggest that a cassoy-urea pellet containing CH and SBM or urea could be used as a protein source in concentrate mixtures for ruminants to improve rumen ecology, nutrient digestibility and ruminal fermentation. Considering the overall result, CSU2 could be used to improve rumen ecology, fermentation and digestibility, however further research in feeding trials with milking cows and fattening beef should be conducted.

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