Rumen pH and Ammonia Nitrogen of Cattle Fed Different Levels of Oil Palm (*Elaeis guineensis*) Frond Based Diet and Dry Matter Degradation of Fractions of Oil Palm Frond

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ABSTRACT : Three fistulated Malaysian local bulls were used in a 3×3 Latin square design to determine the effects of different levels of concentrate with oil palm (*Elaeis guineensis Jacq.*) frond (OPF) on rumen pH and NH_3-N concentration, and DM degradability of different fractions of OPF. Three diets namely, 60% OPF pellet and 40% concentrate (Diet 1), 50% OPF pellet and 50% concentrate (Diet 2) and 40% OPF pellets and 60% concentrate (Diet 3) were used. The levels of concentrate in the diets affected rumen pH and NH_3-N concentration. The pH and NH_3-N concentration almost in all hourly samples did not show any difference ($p>0.05$) among the diets except the 6 h and 9 h samples. The highest ($p<0.01$) NH_3-N concentration was obtained on Diet 3 followed by Diet 2 and Diet 1, but there was a slightly higher ($p>0.05$) pH on Diet 1. The NH_3-N concentrations of rumen liquor at 9 h sampling on Diet 1 and Diet 2 were below the critical level (50 mg/liter) required for efficient fermentation of fibrous feeds. The *in sacco* DM degradation of different fractions of OPF was affected by diets. The DM degradation of fractions of OPF was higher on Diet 3, which showed differences ($p<0.01$) with the other diets. It was found that a higher level of concentrate (60%) with OPF gave a higher rumen NH_3-N concentration that increased the DM degradation of OPF fractions. The results showed that OPF could support an efficient rumen function in terms of NH_3-N concentration and pH when ≤50% in the diet. A higher level of OPF (>50%) does not support an efficient rumen fermentation in terms of NH_3-N concentration, and resulted in lower DM degradation values of the fractions. The results suggested that there is a need to supplement additional nitrogen to OPF based diets. (*Asian-Aus. J. Anim. Sci. 2000. Vol. 13, No. 7 : 941-947*)

Key Words : Oil Palm Frond, Rumen pH, Rumen NH_3-N Concentration, *In Sacco* Degradability

INTRODUCTION

Oil palm (*Elaeis guineensis* Jacq.) frond (OPF) is now used in ruminant diets in Malaysia (Dahlan, 1992, 1996; Islam et al., 1998). The yield, nutrient composition and nutritive value of OPF as a ruminant feed have been studied (Dahlan, 1992; Islam and Dahlan, 1997; Islam, 1999). The rate of degradation of a feed on a nutritionally adequate diet is the initial assessment of a fibrous feed for ruminants in terms of availability of nutrients (Preston, 1995). In an earlier experiment (Islam et al., 1997) using a nutritionally adequate diet (ARC, 1984), the DM degradation obtained at the 48 h incubation of the major fractions of OPF (petiole and leaflet) were about 40 g/100 g. The degradability value showed the potential of OPF as a feed for ruminants (Islam, 1999). Preston (1995) suggested a further evaluation of degradability of the ingredient on the ingredient-based diet. Since OPF is considered as a roughage source, it is necessary to determine the degradability of different fractions of OPF.

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The efficiency of a fibrous feed in the rumen ecosystem cannot be characterized by conventional feed analysis (Preston, 1995). Feed intakes of many diets sometimes bear no relationship with the digestibility and the energy value of the diet, but it is much more influenced by supplementation of the diet (Minson, 1990). The rumen NH_3-N concentration is a limiting factor for rumen microorganisms that affects the digestion of a feed, especially for fiber digestion (Perdock, 1987; Ørskov, 1995). Therefore, it is essential to determine if the OPF can support an efficient rumen function or not. The pH and NH_3-N concentration of rumen liquor present a picture of the fermentation activities of a feed in the rumen. Moreover, the level of rumen ammonia indicates if there is an immediate need to provide additional fermentable nitrogen in the diet (Preston, 1995). Therefore, determinations of rumen pH and NH_3-N concentrations of cattle fed on different levels of OPF in diets were carried out. The objectives of the experiment were to evaluate the effectiveness of the OPF in rumen digestion. The rate and extent of DM degradation of the fractions in the OPF based diets were also determined.

MATERIALS AND METHODS

Animals

Three Malaysian Local (Kedah-Kelantan) bulls each fitted with a 40 mm diameter permanent rumen fistula were used to carry out the nylon bag technique as described by Bhargava and Ørskov (1987). Animals were kept in individual pens. The animals had a 14-day adjustment period prior testing each diet.

Diets

Three different diets based on different levels of OPF were prepared. The diets were: 60% OPF pellet+40% concentrate mixture (Diet 1), 50% OPF pellets+50% concentrate mixture 50% (Diet 2), and 40% OPF pellet+60% concentrate mixture (Diet 3). The concentrate mixture was composed of palm kernel cake (20.0%), rice bran (30.0%), crushed corn (10.0%), soybean meal (35.0%), molasses (3.0%), vitamin-mineral premix (1.5%), and common salt (0.5%). Oil palm frond pellet was offered mixed thoroughly with concentrate mixture in the mangers. Diet was offered twice daily at 08:00 am and 16:00 h. Drinking water was supplied *ad libitum*.

Collection and preservation of rumen liquor

Two hundred ml of rumen fluid from each animal was collected on the 14th day of each trial following the time schedule 0, 2, 4, 6, 9 and 12 h after morning feeding. Samples of rumen fluid were taken using a tube with vacuum pump through fistula. The fluid samples were strained through 4 layers of cheesecloth, and then were preserved following the method of Han et al. (1989). Collected rumen samples were placed in a 250 ml Erlenmeyer flask and acidified with 25% metaphosphoric acid (1 part acid and 5 part rumen liquor). Then the flask was stored at -20℃ until analyses were carried out.

Determination of pH and NH_3-N of rumen fluid

Rumen pH was measured immediately after collection by Corning electronic pH electrode. The NH_3-N concentration of rumen fluids was determined by direct distillation and titration was carried out by an automatic N analyzer [Tecator KJELTEC 1030] (AOAC, 1984).

In sacco DM degradation

Bags measuring 8×13 cm, made of nylon filler cloth stitched with polyester thread (5 stitches per cm) with pore sizes of 20 to 40 μm were used to incubate the samples. Freshly harvested OPF were collected, chipped and separated into petiole, leaflet, and the whole OPF. Samples were then oven dried (60℃ for 72 h), milled through a 4-5 mm screen and kept in pillboxes. Approximately 3 g of each representative sample was placed in each bag. Bags contained the test sample were anchored to a 35 cm plastic tube and incubated in the rumen of each bull. Bags were withdrawn from the rumen at 8, 16, 24, 48, 72 and 96 h of incubation. After withdrawal the bags were washed under running water until the water turned clear, and were then gently squeezed to remove excess water. Bags were then dried at constant 60℃ until constant weight. Water soluble fractions (washing losses) were determined in duplicate by soaking bags with test samples in warm distilled water at 39℃ for 1 h followed by washing and drying as described by Bhargava and Ørskov (1987).

The values for DM degradation at each incubation were mathematically evaluated and fitted to the exponential equation developed by Ørskov and McDonald (1979) and McDonald (1981) to calculate the rate and extent of rumen digestion. The calculation was made using NAWAY-NEW program. The exponential equation is: $p=A+B(1-e^{-ct})$, where, p is the degradation at time t. The lag time was estimated by fitting the model p=A for $t=t_0$, and $p=A+B(1-e^{-ct})$ for $t>t_0$ and the degradation characteristic of the OPF fraction was defined as: the intercept '*A*'=the washing loss (representing the soluble fraction of feed or the rapidly degradable fractions; '*B*'=$(A+B)-A$, is the insoluble but potentially degradable in time t; '*c*' is the rate of degradation of B; and the lag phase (L)=$1/c\ \log_e[B/(A+B)-A]$ (Kibon and Ørskov, 1993).

Statistical analyses

The rumen parameters, the rate and extent of DM degradation of different fractions, were analyzed statistically using analysis of variance in a 3×3 Latin square design with degrees of freedom partitioned to animal, diet and fraction, assuming no carryover effect for each trial (Gomez and Gomez, 1984). The least significance difference (lsd) test of the treatment means ($p<0.01$ and $p<0.05$) was used to compare the diets. The statistical analyses were carried out with the aid of SAS (1997).

RESULTS

Rumen pH

Table 1 shows the effects of different levels of OPF and concentrate on rumen pH. The pH at different hour samples and the mean pH level of the three OPF based diets did not show any significant differences. The mean pH of the Diet 2 was 7.04 which was slight higher compared to Diet 1 (7.01) and Diet 3 (6.99). The mean pH for all diets at 2, 4, 6 and 9 h sampling were almost similar but the values were higher at 0 and 12 h sampling. However, the rumen pH of all the hour samples of the three diets ranged from 6.82 to 7.74.

NH_3-N concentration

The mean NH_3-N concentration was the highest ($p>0.05$) on Diet 3 followed by Diet 2 and Diet 1.

Table 1. The pH and NH_3-N (mg/litre) concentration of rumen fluid of cattle fed on diets prepared from different levels of oil palm frond

Hours	Diets				lsd	Significance level
	1	2	3	Mean		
pH						
0	7.74	7.78	7.68	7.73	0.203	NS
2	7.04	6.91	6.85	6.93	0.343	NS
4	6.93	6.95	6.83	7.90	0.256	NS
6	6.94	6.89	6.86	6.89	0.462	NS
9	6.96	6.84	6.82	6.87	0.462	NS
12	7.03	6.90	6.89	6.94	0.213	NS
Mean	7.10	7.05	6.99	7.05	0.277	NS
NH_3-N						
0	152	164	183	166	84	NS
2	108	128	145	127	81	NS
4	64	100	110	91	87	NS
6	35	58	88	60	20	0.01
9	19	38	55	37	26	0.05
12	40	53	67	53	43	NS
Mean	70	90	108	89	43	NS

NS=Non-significant; Diet 1, 60% OPF and 40% concentrate; Diet 2, 50% OPF and 50% concentrate; Diet 3, 40% OPF and 60% concentrate.

Table 2. Dry matter degradation of petiole, leaflet and the whole oil palm frond (g/100 g) of at different hours of incubation in the rumen of cattle fed on diets prepared from different levels of oil palm frond

Fractions	Diets	Incubation hours (h)					
		8	16	24	48	72	96
Petiole	1	20.98	24.72	30.15	34.03	35.50	36.91
	2	19.32	24.01	31.51	34.25	35.58	37.57
	3	22.42	26.51	31.31	35.76	36.68	39.30
Mean of the diets		20.91	25.08	30.99	34.68	35.92	37.92
Leaflet	1	19.06	21.37	26.50	29.81	32.17	34.33
	2	18.50	22.25	25.76	28.99	31.04	33.41
	3	19.75	23.74	27.65	26.64	34.47	35.85
Mean of the diets		19.10	22.45	26.64	28.48	32.56	34.53
Whole OPF	1	19.33	21.53	26.97	30.20	31.92	33.10
	2	19.24	21.52	25.85	29.71	30.91	33.94
	3	19.37	24.98	28.44	30.85	34.14	35.70
Mean of the diets		19.31	22.67	27.10	30.25	32.32	34.24
lsd and significance level							
Animal		1.4NS	0.72NS	3.55NS	5.25NS	3.3NS	2.5NS
Diets		1.4NS	0.72**	3.55NS	5.25NS	3.3NS	2.5NS
Fractions		1.4NS	0.72**	3.55*	5.25*	3.3**	2.5*

NS=Non-significant; * $p<0.05$, ** $p<0.01$. Diet 1, 60% OPF and 40% concentrate; Diet 2, 50% OPF and 50% concentrate; Diet 3, 40% OPF and 60% concentrate.

The NH_3-N concentration increased correspondingly with increasing level of concentrate in the diet, and the highest was on Diet 3. The mean NH_3-N concentrations for Diet 1, 2 and 3 were 65.0, 83.3 and 98.7 mg/liter, respectively. The NH_3-N concentration decreased linearly from 2 h up to 9 h post-feeding. The 6 and 9 h NH_3-N concentrations were the least in all diets (table 1).

In sacco DM degradation

The DM degradation of the whole OPF and the fractions of OPF in different OPF based diets are presented in table 2. Higher ($p>0.05$) degradation values showed in the petiole compared to leaflet and

Table 3. Digestion characteristics (g/100 g) of dry matter of different fractions of oil palm frond at different levels of the oil palm frond based diets according to the equation, $p=A+B(1-e^{-ct})$

Fractions	Diets	A	B	(A+B)	c	L	RSD
Petiole	1	16.33	20.50	36.83	0.0455	1.83	1.03
	2	13.34	23.52	36.86	0.059	2.50	1.77
	3	16.17	22.67	38.84	0.0429	0.67	1.25
Mean		15.28	22.32	37.50	0.0491	1.67	1.35
Leaflet	1	15.66	21.97	37.63	0.0259	0.30	1.40
	2	14.96	19.13	34.09	0.0318	0.70	1.27
	3	15.00	21.93	36.93	0.0375	1.87	0.62
Mean		15.21	21.68	36.21	0.0317	0.96	1.10
	1	15.27	19.37	34.64	0.037	0.97	1.41
Whole OPF	2	15.79	19.38	35.17	0.0269	0.87	1.39
	3	14.92	21.73	36.65	0.0417	1.36	1.23
Mean		15.30	20.18	35.48	0.0352	1.07	1.36
lsd and significance level							
Animals		2.22NS	3.89 NS	2.86 NS	0.02NS	2.49NS	0.77NS
Diets		2.22NS	3.89 NS	2.86 NS	0.02NS	2.49NS	0.77NS
Fractions		2.22NS	3.89 NS	2.86 NS	0.02*	2.49NS	0.77NS

NS=Non-significant; * $p<0.05$, NH_3-N, Ammonia nitrogen concentrations of the rumen liquor. Diet 1, 60% OPF and 40% concentrate; Diet 2, 50% OPF and 50% concentrate; Diet 3, 40% OPF and 60% concentrate.

whole OPF at all levels of incubation period and all the diets. Highest degradation values of the fractions of OPF were for Diet 3.

Digestion characteristics measured from the exponential equation are presented in table 3. The rapidly disappearing fraction (A) of the fractions of OPF were almost similar ($p>0.05$). Among the diets, A value was slightly higher on Diet 1, followed by Diet 2 and Diet 3. Among the fractions, A values were almost similar (15.28 to 15.30) and the whole OPF showed a slightly higher ($p>0.05$) than fractionates of OPF. The insoluble but potentially degradable fraction (B) was within a narrow range among the fractions of OPF. The potential degradable fractions (A+B) of the fractions of OPF are also presented in table 3. The A+B was negligibly higher ($p>0.05$) in petiole followed by leaflet and whole OPF. Among diets, slightly higher (A+B) values showed on Diet 3 followed by Diet 2 and Diet 1. There were non-significant differences in rate of degradation (c) between diets, but the rate of degradation was higher ($p<0.02$) in the petiole compared to the whole OPF and leaflet among fractions.

DISCUSSION

The objective of this study was to determine if the OPF could support efficient rumen function for digestion. The pH values with all diets were lower ($p<0.01$) compared to the pH values of the nutritionally adequate diet used in an earlier experiment (Islam, 1999). The pH on the nutritionally adequate diet (ARC, 1984) used in the earlier experiment was 7.32. The lower pH values could be due to using pelleted OPF and concentrates. Generally pelleted roughage showed a lower pH level than coarse roughage (Ørskov and Ryle, 1990). They also reported that with ground or pelleted roughage the depression in pH was higher than with long roughage presumably due to greater saliva secretion with the latter. A slightly higher pH in the Diet 1 might be due to a higher level of OPF compared to other diets. The pH values obtained from this experiment were little different from the normal pH level with fibrous feed-based diets (Wanapat and Wachirapakorn, 1990) and also similar to the rumen pH values of cattle fed on rice straw and pasture grass based diet (Chowdhury and Huque, 1997). However, the obtained pH values in this experiment were always being maintained at a higher level than above the critical level of rumen pH 6.0 for fiber digestion (Ørskov and Ryle, 1990). They also reported that the cellulolytic bacteria need a rumen pH between 6.2 to 7.0 and the obtained pH values of the OPF based diets were in the range from 6.2 to 7.0.

The NH_3-N concentration of rumen liquor fed on diets prepared from OPF was lower compared to that in cattle fed on the nutritionally adequate diet prepared following ARC (1984). This could be due to the level and quality of concentrate used in the diet. The 6 and 9 h NH_3-N concentrations of Diet 1 and Diet 2 were below the critical level and those were significantly lower ($p<0.01$ and $p<0.05$, respectively) than that of the Diet 3. In the balanced diet, although the values were above the critical level, the NH_3-N concentration values were lower in 6 and 9 h samplings compared to other sampling period (Islam, 1999). The critical level NH_3-N concentration is that which might affect fiber digestion of a feed (Ørskov and Ryle, 1990). However, the mean NH_3-N mg/litre (6 samples of the scheduled period) of all the diets prepared from OPF were higher than the critical NH_3-N level of rumen liquor of 50 mg/litre (Preston, 1986; Leng, 1990). This suggests that the OPF could support rumen function in terms of NH_3-N production. Highest ammonia N levels at all the samplings were on the Diet 3, where a higher level of concentrate was used. This suggests that using OPF with a proportion of concentrate can increase rumen ammonia level and support fiber digestion. This supports Wanapat and Wachirapakorn (1990) who reported that an increased level of concentrate in rice straw showed a higher NH_3-N concentration in the rumen.

On most diets based on fibrous feed, the primary limitation to the growth of rumen microorganisms is probably the NH_3-N concentration. This must be above the critical level (50 mg/liter) for a considerable period of the day (Sattar and Slyter, 1974). They also reported that a deficiency of NH_3-N concentration resulted in a reduction in rumen microbial populations. Once the pool size of microorganisms decreases, digestibility of the fibrous feed decrease and intake will fall (Ørskov and Ryle, 1990). The higher level of concentrate resulted in a higher NH_3-N concentration in the rumen. The higher level of OPF in the diet resulted in a lower NH_3-N concentration. The NH_3-N concentration at 2 h sampling was higher in all the diets, and might be due to immediate effects of feeding on the rumen environment. Higher concentrations at 2 and 9 h occured in cattle fed on pasture grass and concentrate based diets also given at 08.00 h and 16.00 h (Islam, 1999). All the diets prepared from OPF were pelleted and the animals consumed them within an hour. Preston (1995) reported that 4 and 6 h (post-feeding) were the critical time to measure the NH_3-N concentration. He also mentioned that NH_3-N level must be above the critical level for prolonged periods in fibrous diets, which are digested slowly. Data showed that the NH_3-N concentration in the Diet 1 was not maintained above the critical level of 50 mg/liter for a long period. However, in Diet 2 and Diet 3 NH_3-N concentrations were always above the critical level except 9 h post feeding for Diet 2.

The ideal concentration of rumen NH_3-N for an efficient digestion has been variously estimated at 50-70mg/litre (Satter and Slyter, 1974) and 150 to 200 mg/litre (Krebs and Leng, 1984; Preston, 1986). Boniface et al. (1986) reported the optimal rumen NH_3-N concentrations were from 45 to 120 mg/litre but the value of Perdok (1987) was about 200 mg/litre. The obtained values from the OPF based diets were higher than the reported value of Satter and Slyter (1974) but lower than the reported value of Krebs and Leng (1984) and Perdok (1987).

Irrespective of the fractions and incubation hours the mean degradation values of the fractions of OPF on the nutritionally adequate diet (Islam, 1999) were higher compared to the degradation values of the fractions on the OPF based diet obtained in this study. Considering all the fractions, the rapidly degradable fraction (A) was higher ($p<0.01$) in the nutritionally adequate diet compared to all OPF based diets (tables 2 and 3). This potential degradable fraction (A+B) was highest in leaflet followed by petiole and whole OPF in the earlier experiment (Islam, 1999), but on the diets prepared from OPF the values were higher in petiole compared to leaflet. The potential degradable fraction in earlier experiment (Islam, 1999) was also higher than that of the OPF feed based diet. However, among the OPF based diets, the highest ($p>0.05$) A+B value was on Diet 3 when a higher level of concentrate was given. A higher ($p>0.05$) degradation rate was observed on Diet 3 when a higher level of concentrate was given that showed higher degradation characte- ristics of the fractions of OPF. This further showed that the diets affected the rumen environment as well as rumen degradation characteristics. Diet 3 contained a higher dietary CP value compared to Diet 1 and Diet 2. This supports Reddy (1996) and Pritchard and Males (1985) who reported that increased dietary nitrogen and addition of sufficient fermentable nitrogen to the host animal diets provide required nutrient for microorganisms and consequently increase in the fiber digestion.

Among the three diets, the diet contained 40% OPF and 60% concentrate showed better degradation values and digestion characteristics of the fractions of OPF than those on the other diets. It was also observed that the diet containing 50% OPF and 50% concentrate showed slight higher degradation values than those on Diet 1 when the OPF level was higher (60%). Comparing these values with an earlier study with a nutritionally adequate diet (Islam, 1999), it can be seen that the OPF fractions showed lower degradability values of the fractions on the OPF based diets in this study. The digestion characteristics (*A*, *B* and *c*) in different NH_3-N concentration levels showed differences, and Diet 3 which showed a higher NH_3-N concentrations resulted in higher digestion characteristics. Similar results were found in the earlier study (Islam, 1999) where the additional nitrogen was supplied by fishmeal and crushed khesari (*Phaseolus mungo*), and required minerals were also provided. The level of concentrate supplement and quality of concentrate have a direct effect on an rumen NH_3-N concentration and eventually increase fiber digestion in the rumen.

CONCLUSION

The results showed that OPF can support an efficient rumen function in terms of NH_3-N concentration when 50% or more is used in the diet, but the latter requires additional fermentable N. This result also indicated that OPF can be fed directly to the ruminants, but qualitative improvement may increase the level of OPF in the diet. Higher degradability values on the diet where dietary protein level was higher. This is evidenced that the quality of the diet effects the rumen environment of the fistulated cattle as well as fiber degradation.

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