

EFFECT OF CONCENTRATE TYPE ON DIETARY NITROGEN PARTITION IN LACTATING FRIESIAN HOLSTEIN CROSSBREED COWS

Budi Prasetyo Widyobroto, Çuk Tri Noviandi, Ali Agus, Hari Hartadi¹

ABSTRACT

This research was conducted to determine the effect of different concentrate type on dietary nitrogen partition in lactating Friesian Holstein crossbreed cows (FHC). Four lactating Friesian Holstein crossbreed cows were fed four different concentrate types and degradation rates, namely rapidly degraded starchy concentrate (PC); slowly degraded starchy concentrate (PL); rapidly degraded fibrous concentrate (SC); slowly degraded fibrous concentrate (SL). This research was done in four periods using 4 x 4 Latin Square Design. All animals were fed King grass (*Pennisetum hybrid*) *ad libitum* as a basal diet and concentrate, with roughage to concentrate ratio of 60 : 40. Data was analyzed using variance analysis, followed by Duncan's Multiple Range Test to determine effect of treatment and orthogonal contrast analysis to determine effect of concentrate types and degradation rates. Results on parameters of ruminal fermentation showed that ruminal pH for SL was higher ($P < .05$) than that of PC, PL and SC at all sampling time. The effect of concentrate types on ruminal pH was significant difference at 2, 3, 4 h post feeding, meanwhile effect of concentrate degradation rate was significant difference at 1, 2, 3, 4, 6, 8 h post feeding. Ruminal NH_3 for PL was higher ($P < .01$) than that of PC, SL and SC at 1 h post feeding, meanwhile SL was higher ($P < .05$) than of PL, SC and PC at 3, 4 6 h post feeding. Organic matter degradation in the rumen of PC was higher ($P < .01$) than that of PL, SC and SL at all incubation time. Milk N secretion and N retention were not significantly affected by the treatments. N secretion for PC, PL, SC and SL were 27.8, 18.5, 21.1, and 129.6 g/head/day respectively, meanwhile the N retention were 74.8, 74.3, 96.6 and 62.9 g/head/day respectively. The efficiency of dietary N utilization was not significantly difference among treatments, but PC concentrate tended to be the most efficient.

Key words: Friesian Holstein cows, Concentrate type, Nitrogen partition, Nitrogen efficiency

INTRODUCTION

To meet the energy demand for high-yielding dairy cows in early lactation adequate supply of concentrate is needed. For dairy cows, carbohydrates are important precursors of energy yielding nutrients either concentrate as structural carbohydrates or as non-structural carbohydrates. De Visser *et al.* (1980) were among the first to report differences in ruminal behavior between non-structural carbohydrates, i.e. like starch and sugars, and structural carbohydrates, i.e. like (crude) fiber in dairy concentrates. When large amounts of starch and sugars were included in the concentrates to feed dairy

cows, ruminal pH could be dramatically reduced with a concomitant equally dramatic rise in the levels of propionic, and sometimes lactic acid in rumen fluid. Tamminga *et al.* (1990) presume that structural carbohydrates in ground concentrates probably also play an important positive role in the stabilization of rumen fermentation whereas large amounts of non-structural carbohydrates sometimes have a negative influence. Further research showed that the negative influence of non-structural carbohydrates on rumen fermentation is largely caused by easily degradable carbohydrates such as soluble sugars and rapidly degraded starches (Tamminga unpublished results).

¹ Faculty of Animal Husbandry, Gadjah Mada University, Yogyakarta, Indonesia

The proportion of carbohydrates escaping degradation in the rumen is to a large extent dependent on rate of degradation. Escape of structural carbohydrates is undesirable because the rumen is by far the most important compartment where structural carbohydrates can be degraded. For the host animal, structural carbohydrates which are not degraded in the rumen are largely bulk which has to be cleared from the intestinal tract at the expense of sloughing of intestinal cells giving rise to the loss of considerable amounts of endogenous protein (NRC, 1989).

Rate of degradation of non-structural carbohydrates is important in various ways. First of all it determines the rate at which energy becomes available for microbial growth, often a limiting factor for microbial activity in the rumen which in turn is believed to be important with regard to rumen capacity (Tamminga *et al.*, 1990). Rate of degradation of non-structural carbohydrates also influences the fermentation pattern which in turn determines the amount of energy, which is extracted by the microbes. Contrary to structural carbohydrates, solubility of non-structural carbohydrates is often resulting in a high proportion being degraded very rapidly. In addition, degradation rate shows a wider variation than that of structural carbohydrates (Tamminga *et al.*, 1990).

Fecal N, come from the dietary N which was undegraded in the rumen, its values depend on degradation rate and retention time of feed in the rumen (Soedomo *et al.*, 1996). Tamminga *et al.* (1990) showed that high excretion of N was caused by low degradation of carbohydrates. It caused undegraded protein was excreted in feces with endogen N so that N content in feces will be higher (Ørskov, 1992). De Visser *et al.* (1990) reported that feeding starches, particularly less rapidly degradable starches, has an influence on milk composition. It reduces milk fat but enhances milk protein percentage. The nature of concentrate would affect the pattern of dietary nitrogen partition. Objectives of this research were to determine the effect of different nature of concentrate on

parameters of rumen fermentation and dietary nitrogen partition in lactating FHC cows.

MATERIALS AND METHODS

Animals and Management

Four lactating Friesian Holstein Crossbreeds fitted with a rumen cannula (average initial body weight 348 kg) were used in a 4 x 4 Latin square design. At the beginning of the experiment, animals were in their second to sixth months lactation with milk production range from three to six liters/day. The cows were housed in individual pens, and were fed twice a day at 08.00 and 17.00 h.

Design and Treatments

The experiment was carried out in 4 periods, and each period consisted of 15 days for adaptation followed by 7 days collection period. Animals were fed King grass as basal diet and concentrate (60 : 40). Four concentrates were prepared based on their type ('starchy' P and 'fibrous' S) and its degradation rate ('slowly' L and 'rapidly' C). Degradation rate values was 'dBO8' confirmed from organic matter (OM) loss values in eight hour in situ incubation in the rumen (Soedomo *et al.*, 1997). The type of concentrates were :

- Rapidly degraded starchy concentrates (dBO8 = 0.90) : cassava (PC)
- Slowly degraded starchy concentrates (dBO8 = 0.58) : polished corn (PL)
- Rapidly degraded fibrous concentrates (dBO8 = 0.63) : soybean hull (SC)
- Slowly degraded fibrous concentrates (dBO8 = 0.41) : beer dregs (SL).

Concentrates were calculated at iso-energy and iso-nitrogenous. The ration was calculated based on the animal's body weight and according to NRC recommendation (1989). Ingredient used and chemical composition of the concentrates is shown in Table 1.

Table 1. Ingredient and chemical composition of the concentrates

Item	Concentrates*			
	PC	PL	SC	SL
Ingredient composition :				
Cassava	75.0	-	-	-
Polished corn	-	75.0	-	-
Soybean hulls	-	-	75.0	-
Beer drags	-	-	-	75.0
Rice bran	-	13.2	12.5	11.8
Soybean meal	11.6	-	5.5	5.8
Cottonseed hulls	7.6	5.0	-	-
Molasses	2.3	3.4	4.4	4.7
Urea	2.6	2.5	1.7	1.8
Minerals	0.9	0.9	0.9	0.9
Chemical composition :				
Organic matter(%) ^a	95.51	95.41	91.94	90.60
Crude protein (%) ^a	13.13	13.27	13.22	13.64
NEI (kcal/kg BK) ^c	1394.00	1462.00	1479.00	1326.00
Degradation rate (dBO8. %) ^d	90.00	58.00	63.00	41.00
NDF (%) ^b	14.83	24.48	55.50	71.10
ADF (%) ^b	5.28	6.90	22.67	24.20

^a Proximate analysis at Animal Feed Laboratory, Faculty of Animal Science Gadjah Mada University.

^b Van Soest analysis at Animal Feed Laboratory, Faculty of Animal Science Gadjah Mada University.

^c Calculated based on NRC recommendation (1988).

^d Soedomo *et al.* (1997).

*PC = Rapidly degraded starchy concentrate, PL = Slowly degraded starchy concentrate, SC = Rapidly degraded fibrous concentrate and SL = Slowly degraded fibrous concentrate.

Sampling and analysis

Experimental cows were adapted with their ration for 15 days. Chopped King grass was offered before concentrates were given. For each period the grass were sampled every three days whereas refusal rations were weighed and sampled 200 g every morning. Samples collected were grouped for each treatment for DM and N analysis.

Feces and urine were collected for seven days at the end of each period. Feces were collected for 24 hours, and 2% by weighed was collected for samples. Samples were dried and grouped for each treatment for DM and N analysis. Urine was collected for

24 hours into container in which 200 ml of 10% H₂SO₄ was added. Urine was weighed and 0.5% collected, and sub samples then was frozen and stored at -20°C for N analysis.

Milk productions were recorded every day. Milk samples were taken twice for each period (the day before and the day after collection period) for N analysis.

Samples of the ration, refusal and feces were analyzed for organic matter (OM) and crude protein (CP) according to AOAC procedures (1975); and for neutral detergent fiber (NDF) and acid detergent fiber (ADF) according to Van Soest procedures (1994).

Calculations

Nitrogen absorption was calculated as N intake minus fecal N. Nitrogen retention was calculated as N intake minus fecal N urinary N and milk N.

$$\text{N absorption} = \text{N intake} - \text{fecal N}$$

$$\text{N retention} = \text{N intake} - (\text{fecal N} + \text{urinary N} + \text{milk N})$$

Efficiency of N utilization was calculated as N retention plus milk N equal to N intake.

$$\text{N Efficiency} = \frac{\text{N retention} + \text{milk N}}{\text{N intake}} \times 100\%$$

In this experiment, efficiency of N utilization was calculated according to N which not excreted in feces and urine were used by animals for maintenance, growth and milk production.

In Situ Measurements

Nylon bags, approximately 7 cm x 11 cm, with pore size of 46 μm was filled with approximately 3 g forage and 4 g concentrate (PC, SC, PL, SL). Nylon bags for a given incubation time were tied together with nylon string. The nylon bags were placed in the rumen of each cow at incubation times of 2, 4, 8, 16, 24, and 48 h (in 3 replicate). After incubation, all nylon bags were washed with tap water for 9 min. Bags were then dried at 80 °C for 48 h in oven, desiccated and weighed. Organic matter disappearance from each sample was calculated for each incubation time.

Statistical Analysis

All data were analyzed as a 4 x 4 Latin square design using the following general linear models procedure of SAS (SAS, 1987) and followed by Duncan's new multiple range test. The effect of concentrates types and concentrate degradation rates were determined by orthogonal contrast analysis. Contrasts were: PC PL vs SC SL; PC SC vs PL SL; PC SL vs PL SC

RESULTS AND DISCUSSIONS

Table 2 showed that ruminal pH was affected by treatments at every sampling time.

Minimum pH for starchy concentrate and fibrous concentrate were reached at 4 and 5 h posts feeding respectively (Figure 2). The minimum of pH value for PC, PL, SC and SL were 5.97; 6.32; 6.27 and 6.56, respectively, in which these results were relatively higher than results in the temperate reporting by Widyobroto (1992).

Decreasing of pH in starchy concentrate (PC and PL) compared with fibrous concentrate (SC and SL) could be related with the higher content of soluble carbohydrates and the higher propionic acid proportion in the rumen. This result is coherent with previous report (MC Carthy *et al.*, 1988; Thomas *et al.*, 1986; Herrera-Saldana *et al.*, 1990) that substitution of fibrous concentrate with starchy concentrate in ration reduced ruminal pH.

Ruminal NH_3 kinetic are shown at Table 3 and Figure 2. All concentrates had different response curve begin 1 h post feeding. Ruminal NH_3 are the end product of protein fermentation by rumen microbe. The NH_3 concentration depends on amount of protein degraded in the rumen. Ruminal NH_3 concentration peaked 1 h post feeding for all treatments. The highest NH_3 concentration was PL followed by PC, SC and SL. This evident could be explained that degradation of protein content was higher than other treatments. The ruminal NH_3 concentration was affected by carbohydrate type at 1, 4 and 6 h post feeding, and was affected by degradation rate at 1, 2, 3, 4 and 6 h post feeding.

The ruminal NH_3 concentrations in Table 4 are above minimum requirement for microbial growth i.e. 2 – 5 mg/100 ml (Satter and Slyter, 1974). Mc Donald *et al.* (1987) suggested that ammonia is one of the components for microbial nitrogen synthesis. Based on those results, it was concluded that N was not a limiting factor for microbial nitrogen synthesis in the rumen.

In situ OM degradation estimates are shown in Table 4 and Figure 3. The organic matter degradation was affected ($P < .01$) by treatments at all time incubation. PC concentrate was higher than PL, SC and SL. The highest OM degradation in PC

Table 2. Effect of concentrate types on ruminal pH kinetic post feeding

Time	Concentrate ^a				SE ^b	Contrast ^c		
	PC	PL	SC	SL		12vs34	13vs24	14vs23
08.00	6.75 ^d	6.68 ^{dc}	6.57 ^c	6.76 ^d	0.04	ns	ns	*
09.00	6.58 ^{hi}	6.64 ^h	6.46 ⁱ	6.78 ^g	0.04	ns	**	*
10.00	6.33 ^l	6.52 ^h	6.46 ^{hi}	6.72 ^g	0.05	*	**	ns
11.00	6.10 ^l	6.39 ^h	6.40 ^g	6.65 ^g	0.07	**	**	ns
12.00	5.97 ^l	6.32 ^h	6.27 ^h	6.56 ^g	0.07	**	**	ns
14.00	6.13 ^c	6.34 ^d	6.29 ^{dc}	6.41 ^d	0.06	ns	*	ns
16.00	6.30 ^c	6.46 ^d	6.37 ^{dc}	6.48 ^d	0.03	ns	**	ns

^aPC = rapidly degraded starchy concentrates; PL = slowly degraded starchy concentrates;

SC = rapidly degraded fibrous concentrates; SL = slowly degraded fibrous concentrates.

^bSE = standard error

^c12vs23 = carbohydrate type; 13vs24 = degradation rate; 14vs23 = type x rate

^{d,e,f}Different superscript in the same row showed a significant differences (P<.05)

^{g,h,i}Different superscript in the same row showed a significant differences (P<.01)

concentrate was caused by NDF and ADF content of this concentrate which was lower than that of another concentrate, as presented in Table 1. Organic matter degradation was affected by concentrate types and degradation rate at all time incubation, and it was affected by concentrate types and degradation rate interaction at 2, 4 and 8 h incubation. Widyobroto (1992) suggested that OM degradation in the rumen was affected by proportion of structural carbohydrate and non structural carbohydrate. The starchy concentrate had relative lower structural carbohydrate compared with fibrous

concentrate, so that it showed OM degradation relative higher.

Dijkstra (1994) stated that in the rumen fermentation organic matter produces volatile fatty acids (VFA). Based on that statement, PC concentrate could be predicted for higher VFA production than other treatments.

The result of experiment showed that treatment differences in concentrate type and concentrate degradation rate influence N intake (P<.01). The N intake of fibrous type was higher (P<.01) than the starchy types being 321.2 and 274.6 g/head/day, respectively. These differences were

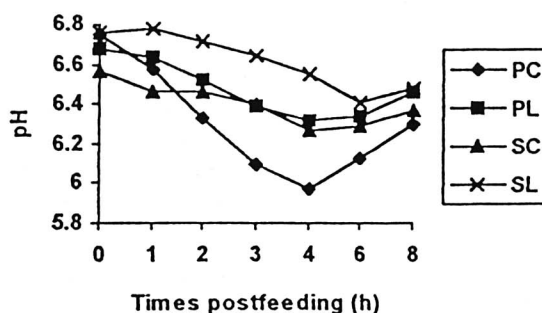


Figure 1. Effect of concentrate types on ruminal pH kinetic post feeding

Table 3. Effect of concentrate types on ruminal NH₃ concentration postfeeding

Incubation time (h)	Concentrate ^a				SE ^b	Contrast ^c		
	PC	PL	SC	SL		12vs34	13vs24	14vs23
08.00	9.34	13.05	8.75	7.67	1.704	ns	ns	ns
09.00	17.42 ^b	21.89 ^a	15.30 ^b	16.95 ^b	1.395	**	**	ns
10.00	15.65 ^b	18.42 ^a	14.73 ^b	16.46 ^{ab}	1.193	ns	**	ns
11.00	10.84 ^b	14.57 ^a	13.91 ^a	15.29 ^a	0.893	*	*	ns
12.00	6.78 ^c	1.29 ^{ab}	11.01 ^b	14.04 ^a	1.003	**	**	ns
14.00	4.20 ^b	7.26 ^a	7.38 ^a	8.56 ^a	1.340	*	**	ns
16.00	3.73	4.92	4.83	5.03	1.021	ns	ns	ns

^aPC = rapidly degraded starchy concentrates; PL = slowly degraded starchy concentrates; SC = rapidly degraded fibrous concentrates; SL = slowly degraded fibrous concentrates

^bSE = standard error

^c12vs23 = carbohydrate type; 13vs24 = degradation rate; 14vs23 = type x rate

^{d,e,f}Different superscript in the same row showed a significant differences (P<.01)

influenced by differences of DM intake (P<.05).

The fecal N losses between PC, PL, SC and SL concentrates was significantly difference but the differences were not showed between PC and PL, PL and SC and between SC and SL concentrates. Statistical analysis showed that fecal N of fibrous types being higher (P<.01) than the starchy types were 79.8 vs 66.1 g/head/day, respectively. Based on N intake, the fecal N between treatments were not significant differences. That non-significant differences were caused by dietary protein solubility which did not differ. Soedomo *et al.* (1997) reported that dietary protein solubility between PC, PL, SC and SL was not significant differences.

The urinary N was significant affected (P<.01) by treatments. The urinary N of PC, PL, SC and SL were 89.5, 132.1, 148.3 and 133.2 g/head/day, respectively. Table 4 showed that urinary N of fibrous type concentrates was higher than starchy types (140.8 vs 110.8 g/head/day), the differences were caused by differences in N intake an available energy for microbe N synthesis on each treatment. Nitrogen excretion by urine was affected by N balance and energy for microbe protein synthesis (Verite and Peyraud, 1988).

The result also indicated that milk N values were non-significant differences between PC, PL, SC and SL concentrate being 27.8, 18.5, 22.1, and 19.6 g/head/day,

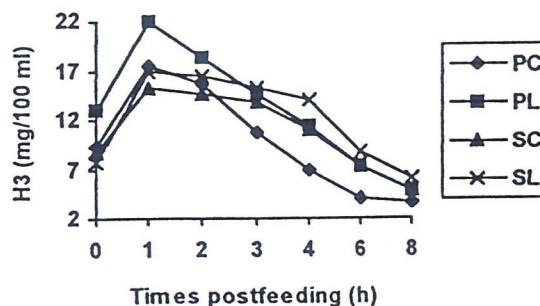


Figure 2. Effect of concentrate types on ruminal NH₃ kinetic post feeding

Table 4. Effect of concentrate types on organic matter degradation in the rumen

Incubation time (h)	Concentrate ^a				SE ^b	Contrast ^c		
	PC	PL	SC	SL		12vs34	13vs24	14vs23
2	42.33 ^d	29.78 ^c	28.31 ^c	25.21 ^f	0.68	**	**	**
4	44.12 ^d	32.01 ^c	30.14 ^c	27.48 ^f	0.71	**	**	**
8	46.25 ^d	37.31 ^c	35.71 ^c	32.63 ^f	0.84	**	**	*
16	51.22 ^d	45.66 ^c	47.00 ^c	42.73 ^f	0.81	**	**	ns
24	56.49 ^d	51.35 ^c	52.68 ^c	49.65 ^e	0.99	*	**	ns
48	64.54 ^d	60.84 ^c	62.77 ^d	58.21 ^f	0.74	*	**	ns

^aPC = rapidly degraded starchy concentrates; PL = slowly degraded starchy concentrates; SC = rapidly degraded fibrous concentrates; SL = slowly degraded fibrous concentrates

^bSE = standard error

^c12vs23 = carbohydrate type; 13vs24 = degradation rate; 14vs23 = type x rate

^{d,e,f}Different superscript in the same row showed a significant differences (P<.01)

or 11.1, 6.4, 6.7, and 6.8% N intake, respectively. Although there is no different, the PC concentrates tended to be increased milk N (27.8 g/head/day). This results are in correspond with reported by Agus (1997) who reported that significant influence of concentrate types on milk protein content that PC concentrate give a higher percentage on milk protein than PL, SC and SL concentrate. Non-significant differences on milk N in this research were presume caused by indifferent amino acid available in the intestine in each treatment so that the amino acid was absorbed not different too.

The treatments with different concentrate types and degradation rates give non-significant effect on N retention. This results were caused by the various effect of

intake N, fecal N, urinary N and milk N values which they complete each other.

The influence of treatment on N efficiency of cows fed PC, PL, SC and SL showed non-significant differences being 40.3, 31.6, 34.6, and 28.3%, respectively. But this efficiency tended to be affected by degradation rates (P=0.056), which the rapidly degradation concentrates the more efficient than the slowly degradation concentrates (37.45 vs 29.95%). Further more, PC concentrates tended to show the most efficient in N utilization (40.3%) than other concentrates. The evidence may be caused by sufficient available energy in PC concentrates for microbe N synthesis, so that dietary protein could be used maximally. Readily available carbohydrates, such as

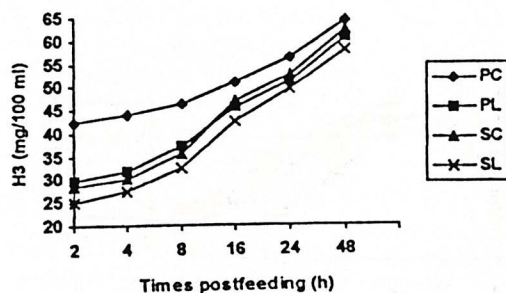


Figure 3. Effect of concentrate types on ruminal pH kinetic post feeding

Table 5. Dietary nitrogen partition

Variables	Concentrates ^a				SE ^b	Contrast ^c		
	PC	PL	SC	SL		12vs34	13vs24	14vs23
	kg/head/day							
DM intake	10.37 ^d	11.07 ^{de}	11.90 ^e	11.48 ^e	0.06	*	ns	ns
	g/head/day							
N intake	254.0 ^p	295.2 ^{pq}	343.6 ^q	298.8 ^{pq}	63.03	**	ns	**
Fecal N	61.9 ^d	70.3 ^{de}	76.5 ^{ef}	83.1 ^f	7.93	**	ns	ns
N absorption	192.1 ^p	224.9 ^{pq}	267.1 ^q	215.7 ^{pq}	66.18	*	ns	**
Urinary N	89.5 ^p	132.1 ^q	148.3 ^q	133.2 ^q	38.26	**	ns	**
Milk N	27.8	18.5	22.1	19.6	13.34	ns	ns	ns
N retention	74.8	74.3	96.6	62.9	102.14	ns	ns	ns
	% N intake							
Fecal N	24.3	23.8	22.3	25.2	0.95	ns	ns	ns
N absorption	75.7	76.2	77.7	72.3	0.97	ns	ns	*
Urinary N	35.4	44.6	43.2	44.1	5.22	ns	ns	ns
Milk N	11.1	6.4	6.7	6.8	2.07	ns	ns	ns
N retention	29.3	25.2	27.9	21.5	7.93	ns	ns	ns
N efficiency	40.3	31.6	34.6	28.3	6.68	ns	ns	ns

^aPC = rapidly degraded starchy concentrates; PL = slowly degraded starchy concentrates; SC = rapidly degraded fibrous concentrates; SL = slowly degraded fibrous concentrates

^bSE = standard error

^c12vs23 = carbohydrate type; 13vs24 = degradation rate; 14vs23 = type x rate

^{d,e,f}Different superscript in the same row showed a significant differences (P < .05)

^{g,h}Different superscript in the same row showed a significant differences (P < .01)

starches and sugars, have been found to be more effective than other carbohydrates for increasing utilization of degraded nitrogen and (or) increasing microbial growth (Offer *et al.*, 1978; Nicholic *et al.*, 1981 cited Varga and Whitsel, 1991). That explanation is equal to Chamberlain and Thomas (1979) statements that the reduce of microbe N synthesis in the rumen could be caused by insufficient energy and less easily fermented carbohydrates. On SC concentrate, it was presume that there was insufficient energy for microbe N activities so that dietary N couldn't optimally used by rumen microbes although N intake on SC concentrate was the highest. In this case, energy, sometimes, to be the limited factor for microbe N synthesis in the rumen (Widyobroto, 1992).

CONCLUSION

Based on the evidence, it was concluded that rapidly degraded concentrates tended to be more efficient in dietary N utilization, especially in PC concentrates. PC concentrates excreted lowest fecal and urinary N, in addition PC concentrate showed the highest N content in its milk than other concentrates.

ACKNOWLEDGEMENT

This experiment was funded by Directorate of Higher Education of Indonesia No:13/P2IPT/DPPM/96/PHB/III/3/1995 May 6, 1996. The authors extend a deep appreciation to Siti Aisyiah, Bambang Wahyudi and Giyarto for technical assistance during the experiment.

REFERENCES

- Agus A. 1997. Pengaruh tipe konsentrat sumber energi dalam ransum sapi perah berproduksi tinggi terhadap produksi dan komposisi susu. *Buletin Peternakan*, Vol. 21 (1):45-54.
- AOAC. 1975. *Official Methods of Analysis*. 12th ed. Association of Official Analytical Chemists. Washington. DC.
- Chamberlain D.G., P.C. Thomas. 1979. Prospective laboratory methodes for estimating the susceptibility of feed proteins microbial breakdown in the rumen. *Proc. Nutr. Soc.*, 38:138A.
- de Visser. H. and A.A.M. de Groot. 1980. The influence of the starch and sugar content of concentrates on feed intake, rumen fluid, production and composition of milk. *Proc. 4th Int. Conf. Prod. Dis.*,:41-48.
- Dijkstra, J. 1994. Production and absorption of volatile fatty acids in the rumen. *Livest. Prod. Sci.*, 39:61-69.
- Herrera-Saldana R., A.R. Gomez, M. Torabi, J.T. Huber, 1990. Influence of synchronizing protein and starch degradation in the rumen on nutrient utilization and microbial protein synthesis. *J. Dairy Sci.*, 73:142-148.
- McCarthy R.D., T.H. Klusmeyer, J.L. Vicini, J.H. Clarck. 1989. Effect of source of protein and carbohydrate on ruminal fermentation and passage of nutrient to the small intestine of lactating cows. *J. Dairy Sci.*, 72:2002-2016.
- McDonald P., R.A. Edwards and J.F.D Greenhalgh. 1987. *Animal Nutrition*. 4th edition. English language Book Society, Longman, London.
- NRC. 1988. *Nutrient Requirements of Dairy Cattle*. 8th ed. National Academy Press. Washington. DC.
- Ørskov. E.R. 1992. *Protein Nutrition in Ruminant*. 2nd ed. Academic Press. New York.
- SAS Institut Inc., 1987. *SAS/STAT Guide for personal Computers, Version 6 Editions*. SAS Institut Inc., Cary, NC. Pp.:1028.
- Satter, L. D. and L. L. Slyter. 1974. Effect of ammonia concentration on rumen microbial protein production in vitro. *Br. J. Nutr.*, 32:199-208.
- Soedomo-Reksohadiprodjo. M. Soejono. H. Hartadi dan B.P. Widyobroto. 1996. Manajemen nutrien sapi perah sebagai kontribusi untuk pencegahan polusi lingkungan. 2. Pengaruh aras konsentrat dalam ransum basal rumput King Grass terhadap pencernaan nutrien. sintesis N mikroba dan neraca N pada sapi perah. *Laporan Penelitian Hibah III/2 Perguruan Tinggi*. Fakultas Peternakan. Universitas Gadjah Mada. Yogyakarta.
- Soedomo-Reksohadiprodjo. M. Soejono dan H. Hartadi. 1997. Manajemen nutrien sapi perah sebagai kontribusi untuk pencegahan polusi lingkungan. 3. Pengaruh asal konsentrat dalam ransum basal rumput King Grass terhadap pencernaan nutrien. sintesis N mikroba dan neraca N pada sapi perah. *Laporan Penelitian Hibah III/3 Perguruan Tinggi*. Fakultas Peternakan. Universitas Gadjah Mada. Yogyakarta.
- Tamminga. S.. A.M. Van Vuuren. C.J. Van Der Koelen. R.S. Ketelaar and P.L. Van Der Togt. 1990. Ruminal behavior of structural carbohydrates. non-structural carbohydrates and crude protein from concentrate ingredients in dairy cows. *J. Agr. Sci.*, 38:513-526.
- Thomas C., K. Aston, S.R. Daley and Jaqueline Bass. 1986. Milk production from silage. 4. The effect of the composition of the supplement. *Anim. Prod.*, 42:315-325.
- Van Soest. P.J. 1994. *Nutritional Ecology of The Ruminant*. 2nd ed. Comstock Publishing Associates. A Division of Cornell University Press. Ithaca and London.
- Varga, G.A. and T.J. Whitsel. 1991. Effect of non structural to structural carbohydrates ratio on rate and extent

- of nutrient utilization in situ. *Anim. Feed. Sci. and Tech.*, 32:275-286.
- Verite R., J.L. Peyraud., 1988. Nutrition Azotee. In : *Alimentation des Bovins, Ovins et Caprins*. Pp.:75-93.
- Widyobroto B.P. 1992. Pengaruh aras konsentrat dalam ransum terhadap kecernaan dan sintesis N mikroba dalam rumen pada sapi perah. *Buletin Peternakan Edisi Khusus.*:241-249.
- Widyobroto B.P. 1992. Influence de la proportion et de la nature du concetre sur les sites et la dynamique de la digestion chez la vache haute productrice. *These de du Docteur de l'Universite de Rennes I*. France.