Effect of Dietary Feeding Regimens on Urea and Protein Concentration of Milk in Murrah Buffaloes

B. Roy[1], R. K. Mehla* and S. K. Sirohi[2]

Dairy Cattle Breeding Division. National Dairy Research Institute. Karnal. Haryana-132001. India

ABSTRACT : The present study was planned to examine the effect of different feeding regimens on milk urea concentration and milk protein concentration. The objectives are to describe the diurnal variations of milk urea (MU) concentration and to predict plasma urea (PU) concentration from MU concentration. Six lactating Murrah buffaloes were distributed in two groups and were fed two different diets in a crossover design. The diets consisted of leguminous crops as diet 1 (berseem (*Trifolium alexandrinum*)+concentrate mixture 1+wheat straw)) and non-leguminous crops as diet 2 (oats (*Avana sativa*)+concentrate mixture 2+wheat straw). All the diets were isocaloric and isonitrogenous. Each diet was fed to the animals for a period of 28 days, followed by a 10 day gap to obviate the carry over effect of the previous diet and then a switch over to the other diet. Digestibility trials were conducted on the last 7 days of each feeding period. Milk samples were collected on day 3, 7, 10, 14, 17, 21, 24 and 28 of the feeding period and blood samples were collected on the same day at morning within 30 minutes after morning milking. The average milk urea (MU) values (mg/dl) differed significantly (p<0.01) and were 44.83±0.62 and 42.53±0.73, respectively, for diets 1 and 2. Milk urea concentrations (mg/dl) also varied (p 0.01) among the days of feeding period, but were stabilized after 10th day of feeding period. In contrast, diets and days of feeding period had no significant effect on percent milk protein. Plasma urea concentration showed a significant (p<0.01) positive correlation (r = 0.93) with MU concentration. To predict the PU from MU the following equation was developed "PU = 10.67±0.76×MU (mg/dl) with R^2 = 0.87". A clear diurnal variation of MU was found with lowered morning value (42.04±0.68 mg/dl) than the evening value (45.32±0.66 mg/dl). Present findings suggested that MU or PU concentration could be used as an indicator to monitor the feeding strategy. Plasma urea can be predicted from MU, whenever interpretation of milk urea data required consideration of diurnal variation. *(Asian-Aust. J. Anim. Sci. 2005. Vol 18, No. 7 : 973-979)*

INTRODUCTION

Of all domestic animals, the buffalo holds the greatest promise and potential for milk production. It is the principal dairy animal in India and play very important role in the rural economy. Surprisingly, in India buffalo farming is practiced on non-scientific lines and there is no tradition of consulting animal health, reproduction or nutritional experts in identifying or addressing the relevant problems. Under conventional farming system diets are not formulated according to the requirements of individual animals, affecting overall production and poor health and reproduction.

In recent years, the adequate protein and energy nutrition of dairy animals has been increasingly constrained. A key to efficient feed utilization is to formulate a ration that optimizes microbial protein synthesis and supply amount of rumen undegradable or bypass protein that

provides additional protein to meet milk production requirement. The balance is associated with baseline concentration of urea in plasma and milk. High endogenous concentrations of urea have been associated with impaired fertility, reduced energy availability, environmental pollution concerns and economic losses (Ferguson and Chalupa. 1989; Gooden et al., 2001; Qureshi et al., 2002). Using milk protein and milk urea concentrations in either blood or milk to monitor dietary energy and protein intake in dairy cows has obtained increased interest in Europe (Nagel, 1994), Japan (Ougi. 1994) and United States (Hutjens and Barmore, 1995). The urea concentration in plasma and milk in cattle are influenced by the amount of crude protein in the diet (Carlsson and Bergstrom. 1994; Gonda and Lindberg, 1994; Baker et al., 1995), as well as by degradable intake protein (DIP) and undegradable intake protein (UIP) (Ropstad et al., 1989). Erbersdobler and Zucker (1980) and Oltner and Wiktorsson (1983) have postulated that a surplus of N intake increases blood urea nitrogen (BUN) which has a close relationship with milk urea nitrogen; MUN (Eckart, 1980; Oltner and Wiktorsson. 1983; Dhali, 2001), because urea freely diffuses from blood to milk (Gustafsson and Palmquist. 1993). In turn, BUN is also affected by several factors, including level of CP in the diet (Oltner et al., 1985; Rosler et al., 1993; Baker et al., 1995) and carbohydrate composition (Lykos et al., 1997).

Table 1. Composition of different feedstuffs used during the digestibility trials (on % DM basis)

Periods	Diets	Feed composition	DM	OM	TCHO	CP	EE	CF	NFE	ASH
I[st]	1	Berseem (*Trifolium alexandrinum*)	12.30	87.21	66.54	17.48	3.19	20.03	46.51	12.79
		Wheat straw	89.40	88.42	84.05	3.06	1.31	48.40	35.65	11.58
		Concentrate mixture-1 (wheat -49%, wheat bran-30%, groundnut cake-18%, mineral mixture-2% and common salt-1%)	91.16	93.04	73.59	16.30	3.15	6.75	66.84	6.97
	2	Oats (*Avana sativa*)	20.51	89.46	80.42	8.02	2.38	34.40	46.02	10.55
		Wheat straw	89.40	88.42	84.05	3.06	1.31	48.40	35.65	11.58
		Concentrate mixture-2 (wheat -38%, wheat bran-29%, groundnut cake-30%, mineral mixture-2% and common salt-1%)	90.04	92.38	64.17	23.99	4.20	9.63	54.54	7.64
2nd	1	Berseem (*Trifolium alexandrinum*)	16.64	88.50	70.71	15.80	1.99	32.50	38.21	11.50
		Wheat straw	91.24	87.73	83.93	3.23	1.27	44.53	39.40	12.28
		Concentrate mixture-1 (wheat -49%, wheat bran-30%, groundnut cake-18%, mineral mixture-2% and common salt-1%)	89.78	91.73	72.31	16.48	2.94	7.85	64.46	8.28
	2	Oats (*Avana sativa*)	27.60	89.48	79.60	8.20	2.68	32.13	47.47	10.53
		Wheat straw	91.24	87.73	83.93	3.23	1.27	44.53	39.40	12.28
		Concentrate mixture-2 (wheat -38%, wheat bran-29%, groundnut cake-30%, mineral mixture-2% and common salt-1%)	90.84	92.08	66.40	22.70	3.98	10.33	56.07	7.93

DM = Dry matter, OM = Organic matter, TCHO = Total carbohydrate.

The aim of the present study was to examine the effect of different feeding regimen on milk urea (MU) concentration and milk protein content and to describe the diurnal variations of MU concentration and to predict plasma urea (PU) concentration from MU concentration.

MATERIALS AND METHODS

Experimental design and treatments

Six lactating Murrah buffaloes were selected from buffalo herd of National Dairy Research Institute. Cross over design was used for the experiment. Animals were divided into two groups with three animals in each group. Two different diets, diet 1 (leguminous crop based) and diet 2 (non-leguminous crop based) were formulated which were isocaloric and isonitrogenous. For individual animal diets were formulated according to Kearl Feeding Standard (1982). Two diets were fed to two different groups of the animal for a period of 28 days and then buffaloes were returned to the normal herd feeding for a period of 10 days to obviate the carry over effect of the previous diet. After 10 days buffaloes were switch over to the next diet by interchanging the diets. For diet 1, wheat straw mixed with 50 percent of the total concentrate mixture-1 (required per day) and berseem (*Trifolium alexandrinum*) fodder provided to the animals separately in feeding manger. Oats (*Avana sativa*) fodder and wheat straw were mixed along with 50 percent of the total concentrate mixture-2 (required per day) to prepare diet 2. Both the diets were provided to the animals twice daily at 9.30 to 10.00 a.m. and 5.00 to 5.30 p.m. Remaining 50 percent of the total concentrate

mixture was fed in two parts to individual buffaloes at the time of milking, morning 5.30 a.m. and evening 6.30 p.m. Animals were offered water thrice a day, at morning, noon and evening. Animals were allotted to the different feeding regimens as given in Table 1. In both the diets, ratio of concentrate and roughage was 40:60 and green fodder and straw was 3:1 on DM basis.

Digestibility trial and analytical techniques

The digestibility trials were conducted in last 7 days of each feeding period for both the diets. Samples of feed offered and residues left over, if any, were taken each morning for DM determination and pooled samples of 7 days were analysed for proximate principles. The quantity of faeces voided during 24 h period was recorded in the morning. Dry matter, organic matter, crude protein (N×6.25), ether extract, total carbohydrates and total ash were estimated as per AOAC (1995). Total digestible nutrients (TDN) were calculated by adding the digestible nutrients (crude protein+total carbohydrate+crude fat× 2.25). DE, ME and NE_L was calculated with the help of following formula as per NRC (1989). DE (Mcal/kg of DM) = 0. 04409×TDN (%of DM). ME (Mcal/kg of DM) = -0. 45+ 1.01 DE. NE_L (Mcal/kg of DM) = 0. 0245×TDN (%of DM)-0.12.

Collection of milk and analytical techniques

Milk samples were collected twice a week on the same day of blood collection during each feeding period to determine the urea and protein content. For individual animal, morning and evening milk samples were collected

Table 2. Various nutrients intake from two dietary groups

Nutrients (kg/day)	Diet 1	Diet 2
DM	11.22±0.51	10.84±0.77
CP	1.64±0.07	1.52±0.06
CF	2.52±0.13	2.67±0.22
EE	0.277±0.030**	0.352±0.017**
NFE	5.58±0.30	5.45±0.43
OM	10.06±0.45	9.94±0.67

** Significant at p<0.01.

separately in capped plastic bottle. The samples were collected after complete milking and thorough mixing from the milk weighing bucket. After collection, samples were kept in refrigerator at 4°C and were analysed on the same day for urea and protein. Milk protein was determined by Kjeldahl method (AOAC, 1995). Milk urea concentration was determined by a modified colorimetric DMAB (p-Dimethylaminobenzaldehyde) assay (Bector et al., 1998).

Blood collection and analysis

Blood samples were collected twice a week during each feeding period to determine the plasma urea concentration. The blood was drawn from the jugular vein into heparinized (20 I.U. heparin/per ml blood) at morning between 6.00 to 6.30 am, within 30 minutes after morning milking. Immediately after sampling, the blood was centrifuged at 3,000 rpm for 15 to 20 minutes and the plasma was separated and stored frozen (-20°C) until analysed. The plasma urea was estimated according to Rahmatullah and Boyde (1980).

Statistical analysis

The data were subjected to least squares analysis of variance (Harvey, 1987) using fixed least squares model procedure. The mathematical model for the analysis was as under.

Mathematical model:

$$Y_{ijklm} = \mu + D_i + P_j + T_k + G_l + e_{ijklm}$$

Where,

Y_{ijklm} = Observation of lth group in i^{th} diet, j^{th} day and k^{th} time of milking

μ = Overall mean

D_i = Fixed effect of ith diet (i = 1 and 2)

P_j = Fixed effect of jth day of feeding period (j = 1, 2, 3, 4, 5, 6, 7 and 8)

T_k = Fixed effect of kth time of milking (k = 1 and 2)

G_l = Fixed effect of lth group of animals (l = 1 and 2)

e_{ijklm} = Random error, which is assumed to be normally and independently distributed with zero mean and constant variance $\sigma^2 e$.

The least squares means for different traits at different

Table 3. Digestibility (%) of various nutrients in two dietary groups

Nutrients (%)	Diet 1	Diet 2
DM	72.64±0.44	70.07±0.85
CP	73.31±0.56*	70.75±0.91*
CF	68.88±0.70*	66.36±0.78*
EE	68.43±1.45**	75.76±1.69**
OM	76.30±0.40*	73.35±0.81*
NFE	73.99±0.64*	69.94±1.19*

** Significant at p<0.01. * Significant at p<0.05.

Table 4. Various digestible nutrients intake from two dietary groups

Nutrients (kg/day)	Diet 1	Diet 2
DM	8.15±0.35	7.58±0.47
CP	1.20±0.05	1.08±0.04
CF	1.74±0.09	1.78±0.15
EE	0.192±0.024**	0.265±0.010**
NFE	4.13±0.22	3.81±0.27
OM	7.67±0.34	7.27±0.42

** Significant at p<0.01.

levels were compared by using Duncan's Multiple Range Test (DMRT) as modified by Kramer (1957). For determining the association between the different traits the correlation analysis was performed. To predict the plasma urea concentration (Y) on the basis of urea concentration in milk (X), the regression analysis was performed. The model was as below.

$$Y_i = a + bX_i + e_i$$

Where,

Y = Predicted plasma urea value

a = Constant for milk urea value

b = Regression coefficient

X = Milk urea value

e = Random error, which is assumed to be normally and independently distributed with zero mean and constant variance $\sigma^2 e$.

RESULTS AND DISCUSSION

Intake of various nutrients and concentration of urea and protein in milk

The proximate analyses of different feed ingredients used in two different diets have been shown in Table 1. Nutrient intakes by the animals (kg/day) from the two different diets have been presented in Table 2. Intakes of various nutrients did not differ significantly except ether extract. Ether extract intake was significantly (p<0.01) higher in diet 2 (0.352±0.017) than diet 1 (0.277±0.030). The digestibility (%) of different nutrients and the intake of different digestible nutrient (kg/day) have been presented in Table 3 and 4, respectively. Dry matter (DM) digestibility did not differ significantly between the diets. However, the

Table 5. Protein and energy utilization by the animals from two dietary groups

Particulars	Diet 1	Diet 2
CP intake (kg/day)	1.64±0.07	1.52±0.06
DCP intake (kg/day)	1.20±0.05	1.08±0.04
TDN intake (kg/day)	7.54±0.35	7.24±0.40
DE intake (Mcal/day)	33.24±1.52	31.90±1.76
ME intake (Mcal/day)	28.52±1.31	27.34±1.43
NE_L intake (Mcal/day)	17.12±0.79	16.43±0.89
CP/ME (g/Mcal)	57.68±0.54	55.92±1.19
CP/NE_L (g/Mcal)	96.05±0.87	93.13±2.07
DCP/ME (g/Mcal)	42.28±0.43*	39.61±1.28*
DCP/NE_L(g/Mcal)	70.41±0.74*	65.97±2.20*

* Significant at p<0.05.

Table 6. Effect of diets, days of feeding period and time of milking on milk urea (MU) and milk protein (MP) concentration

Particulars	Milk urea (mg/dl)	Milk protein (%)
Diets		
1	44.83±0.62[a]	3.93±0.04
2	42.53±0.73[b]	3.88±0.05
Days of feeding period		
3	47.53±1.95[a]	4.12±0.14
7	49.18±1.35[a]	3.89±0.07
10	46.62±1.45[a]	3.88±0.10
14	40.81±1.46[bc]	3.74±0.09
17	43.37±0.96[b]	3.98±0.09
21	41.31±0.63[bc]	3.78±0.11
24	40.65±0.59[bc]	3.90±0.07
28	40.10±0.57[c]	3.95±0.06
Time of milking		
Morning	42.04±0.68[a]	3.90±0.03
Evening	45.32±0.66[b]	3.89±0.07

[a,b,c] Superscript indicate values with different superscripts within column differ significantly (p<0.01).

value was higher (72.64±0.44) in diet 1 than diet 2 (70.07±0.85). The crude protein digestibility was 73.31±0.56 and 70.75±0.91 for diets 1 and 2, respectively, and they differ significantly (p<0.05). The digestibility of crude fibre (CF), organic matter (OM) and nitrogen free extract (NFE) also differ significantly (p<0.05) between the diets, where as, ether extract (EE) digestibility varied significantly at p<0.01. The digestible nutrient intakes (kg/day) did not differ significantly, except digestible EE (Table 4). The utilization pattern of CP in relation to energy has been presented in Table 5. The TDN, CP, DCP, ME and NE_L intake did not differ significantly between the diets. The intake of CP and per unit of ME and NE_L were higher in diet 1 than diet 2, but did not differ significantly. Whereas, DCP intake per unit of ME and NE_L were significantly higher in diet 1.

The concentration of MU and milk protein under different diets on different days of feeding period and time of milking are presented in Table 6. The effect of different days of feeding period and diets on MU was found

Table 7. Correlation coefficients of different combinations of milk urea (MU), plasma urea (PU) and milk protein (MP) concentrations

Particulars	Correlation coefficient (r)
Morning milk urea: evening milk urea	0.86**
Morning milk urea: plasma urea	0.93**
Morning milk urea: average daily milk urea	0.96**
Evening milk urea: plasma urea	0.86**
Evening milk urea: average daily milk urea	0.96**
Average daily milk urea: plasma urea	0.93**
Morning milk urea: milk protein content	-0.0135
Evening milk urea: milk protein content	0.13
Average daily milk urea: average daily milk protein content	0.08

** Indicates that the r value is significant at p<0.01.

significant (p<0.01). When all the observations of different days of feeding period and time of milking were pooled, the average concentration (mg/dl) for diet 1 (44.83±0.62) was higher (p<0.01) than diet 2 (42.53±0.73). It was evident that the average MU concentration was low after 10th day of feeding period and almost stabilized after that. However, protein content of milk did not differ significantly between the diets and among the different days of feeding period (Table 6). A weak positive correlation was found between daily MU and MP content (Table 7).

Urea is the primary form of metabolic end product of protein catabolism in the body, which equilibrates rapidly throughout the body fluids, including milk (Gustafsson and Palmquist, 1993). Excess N supplied to the rumen or post ruminal tissues or energy deficient diet results in high endogenous concentrations of urea in blood and milk. The concentrations of urea in blood and milk are readily affected by dietary intake of protein and energy. In our study, significantly (p<0.01) higher MU concentration in diet 1, might be due to a highly degradable protein source i.e., leguminous fodder (berseem) as compared to the non-leguminous fodder (oats) (Cheeke, 1991). Besides this, digestibility of all the nutrients was also higher in diet 1, except EE. Highly digestible forage can only be obtained after high N fertilization, which not only stimulate forage growth and digestibility, but also increased its N content relative to available energy (Hof et al., 1994). Bertoni et al. (1990) suggested that increased urea level in blood and milk is not itself an indicator of protein excess but should be considered in relation to other factors such as type of proteins. The CP:ME and $CP:NE_L$ did not differ significantly between the diets (Table 5). While, DCP:ME and $DCP:NE_L$ were significantly (p<0.05) higher in diet 1 than diet 2. Higher protein: energy ratio contributes higher MU in diet 1. The main factor influencing the MU content was not only the amount of protein ingested in relation to requirement, but also the relationship between the protein and energy in the ration (Oltner and Wiktorsson, 1983;
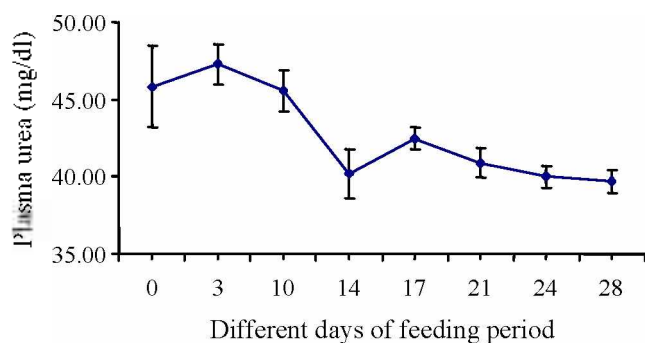
Figure 1. Average plasma urea (PU) concentration of different days of feeding period.

Cheli et al.. 1992; Bertoni et al.. 1993; Whitaker et al., 1995; Dhali. 2001). In diet 2, concentrate, wheat straw and oats (chopped) fodder were mixed and allowed to the animals. as total mixed ration. Whereas. in diet 1. wheat straw mixed with concentrate and berseem fodder provided to the animals separately in feeding manger. Feeding strategy might influence the urea concentration in plasma and milk. The cows fed a total mixed ration had lower ruminal ammonia and plasma urea N concentration compared with cows fed forage and concentrate separately (Carrol et al.. 1988). The average MU concentrations (mg/dl) at different days of feeding period clearly indicated that initially there were higher values. but after 10th day the values were stabilized (Table 6). The results of this study confirm the findings of Oltner and Wiktorsson (1983) who concluded that the change in MU content in response to an altered dietary condition was very rapid and that within a few days the urea level had stabilized at the new concentration.

In this study. no significant differences were found regarding milk protein content between the diets and among the different days of feeding period. although milk urea concentration varied significantly (Table 6). The average milk protein content (%) for diets 1 and 2 were 3.93+0.04 and 3.88+0.05. respectively. The values were within normal range of protein content of buffalo milk. Buffalo milk contains about 3.8% milk protein (Banerjee, 1998; Roy et al., 2003). Urea nitrogen level in milk and blood reflects ruminal protein degradation and post ruminal protein provision. where as milk protein concentration responds to dietary energy intake and bacterial protein production in the rumen (Hwang et al.. 2000). Milk protein reflects the balance of dietary protein and energy intake (Rosler et al., 1993; Sato. 1998). Milk protein yield is to be more highly correlated to nutrient intake than to nutrient composition (Lyatuu and Eastridge, 1998). Coulon and Remand (1991) suggested that milk protein can be used as an indicator for energy intake. because it responds to the energy supply level of the diary cows. In this study, normal range of milk protein indicates that the diets were balanced in terms of energy and protein.

MU values in this study were found in little higher side in comparison to the other studies. most of which were reported in dairy cows. Still the values are comparable with the other studies. MU level in Murrah buffaloes ranged between 25-60 mg/dl as reported by Singhal et al. (1994). Qureshi et al. (2002) found average serum urea level as high as 46 mg/dl in Nili-Ravi Buffaloes. The little higher MU values in this study may be due to the species variation or it may be due to the use of specific feeding standard (Kearl Feeding Standard, 1982). In Germany. the standard milk urea nitrogen value is 7.0 to 14.0 mg/dl (MU value = 16 to 32 mg/dl) (Nagel. 1994). In United States, the value is 11 to 17 mg/dl (MU value = 25 to 38 mg/dl) (Hutjens and Barmore. 1995) and in Japan it is 8 to 20 mg/dl (MU value = 18 to 45 mg/dl) (Ogui, 1994).

Plasma urea and its relation with milk urea

Urea was also estimated in blood plasma, collected within 30 minutes after morning milking. Like MU. plasma urea (PU) concentrations (mg/dl) were lower after 10th days of feeding period and were found to stabilize (Figure 1). PU concentration showed a significant correlation (p<0.01) with MU (Table 7). The correlation of PU with morning MU. evening MU and average daily MU were significant (p<0.01) and 'r' values were 0.93. 0.86 and 0.93. respectively.

PU concentration showed a significant (p<0.01) positive correlation (r = 0.93) with MU (Table 7). Similar results found by other studies (Gustafsson and Palmquist. 1993; Roseler et al., 1993; Baker et al.. 1995; Dhali. 2001). suggested that urea in the blood system is the major source of urea nitrogen in milk. Plasma urea N (PUN) and MUN were positively correlated. probably because of the passive diffusion of urea from plasma to milk (Clark et al.. 1978). When MU was regressed against PU. a linear relationship was determined and expressed by the following equation: PU (mg/dl) = 10.67+0.76×MU (mg/dl) (R^2 = 0.87). which is similar to results reported by other researchers (Roseler et al.. 1993; Gonda and Lindberg. 1994; Campanile et al.. 1998; Dhali. 2001). Gusatafsson and Palmquist (1993) found that the urea in equilibrated with serum with a time lag 1 to 2 h. when the rate of change in serum was 0.5 to 1.0 mM/h. At this rate, average difference between milk and serum urea was 0.8 mmol. In our study. the average plasma urea level was slightly higher than the milk urea by 0.70 mg/dl. The findings are in accordance with the results of Oltner and Wiktorsson (1983) and Dhali (2001).

Diurnal variation

During the experiment. urea and protein concentration in milk were estimated from morning and evening milking. Urea concentration (mg/dl) in the morning milk (42.04±0.68) was significantly (p<0.01) lower than the evening (45.32±0.66). While. non-significant variation was

found between morning and evening milk protein content. The correlation between morning and evening milk urea concentration (Table 7) was 0.86 (p<0.01).

Diurnal variation of urea content in milk may be a potential source of variation among various other factors. In accordance with the report by Gustafsson and Palmquist (1983) and Dhali (2001) there was considerable diurnal variation in the MU concentration. The diurnal variation may be due to the feeding schedule followed during the experiment. Diets were provided to the animal twice daily at 9:30 to 10:00 a.m. and 5:00 to 5.30 p.m. and milking was done at morning 5:30 a.m. and evening 6:30 p.m. The diurnal pattern mainly found with twice daily feeding frequencies. At higher feeding frequencies, a diurnal pattern remained absent (Thomas and Kelly, 1976; Folman et al., 1981). From regular milking interval, MU content was significantly lower in morning milk (Miettinen and Junoven, 1990). Higher MU content of evening milk in our study may be due to the time gap (1 to 1.5 h) between evening feeding and milking. The peak urea content in blood appeared at 2 to 4 h after feeding (Coggins and Field, 1976; Thomas, 1980; Manston et al., 1981). Miettinen and Junoven (1990) also reported that mainly feeding time influenced milk urea concentration and approximately 2 h after the feeding, MU concentration was highest. Rodriguez et al. (1997) found that the plasma and milk urea nitrogen varied throughout the day in relation to the time of feeding. Broderick and clayton (1997) reported that the lower proportion of total urea concentration in milk was observed in a.m. sampling (1.8%) than in the p.m. sampling (3.3%). Several factors should be considered while interpreting milk and blood urea nitrogen data. Such as the diurnal variations, lag time between the peak level of the BUN to that of MUN (Gustafsson and Palmquist, 1983), the different permeability of the mammary duct tissue (Linzell and Peaker, 1971), and the differences in specific gravity between various components of milk solids that could alter the relative concentration of urea in milk (Roseler et al., 1993).

CONCLUSIONS

Feeding regimens had significant effect on milk urea concentration, although diets were isocaloric and isonitrogenous. This effect might be due to the difference in quality and type of protein between the diets and feeding strategy of the experiment. Because of the different feeding strategy, individual farm should fix their own MU value to maintain the nutritional status of the herd. A highly significant correlation was found between MU and PU. Milk analysis has advantageous over blood because of difficulties associated with collection of blood samples under field conditions as compared to milk collection. The

clear diurnal variation of MU concentration indicated that sampling time should be considered during interpretation of MU values.

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