Effect of Concentrate Feeding Frequency versus Total Mixed Ration on Lactational Performance and Ruminal Characteristics of Holstein Cows

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ABSTRACT : This study was conducted to determine a proper feeding regime for lactating Holstein cows during the warm season in Taiwan. In Feeding Trial, 21 lactating Holstein cows were randomly allotted into three feeding regimes in a Completely Randomized Design. The feeding regimes were roughage fed *ad libitum* along with concentrate fed either twice daily (2C) or four times daily (4C), and total mixed ration (TMR) for 8 weeks. No significant differences among the three feeding regimes were found in body weight changes, and intakes in terms of dry matter, crude protein and net energy. For milk yield and 4% FCM yield, 4C were higher than the other two feeding regimes ($p<0.05$). No significant differences were found in milk compositions or differences noted in the yields of the respective milk components. TMR was worse than 2C and 4C ($p<0.05$) in milk production efficiencies in terms of dry matter intake per kg milk yield, crude protein intake per kg milk protein yield, as well as dry matter intake, net energy intake and crude protein intake per kg 4% FCM yield. Three ruminally fistulated Holstein dry cows were randomly assigned into the same three feeding regimes in a 3×3 Latin Square Design. No significant differences were found among the feeding regimes in ruminal pH, ammonia nitrogen, total VFA concentration, molar percentages of VFA, and acetic acid: propionic acid ratio (C_2/C_3). Taken all together, roughage fed ad libitum and concentrate fed four times daily was the better feeding regime for lactating cows during the warm season in Taiwan. ***(Asian-Aust. J. Anim. Sci. 2002. Vol 15, No. 5 : 658-664)***

Key Words : Feeding Frequency, TMR, VFA, Milk Production, Holstein Cow

INTRODUCTION

There are two popular automatic dairy cow feeding systems in Taiwan, i.e., total mixed ration (TMR) and computerized automatic feeding (CAF). TMR was developed in North America during late 1950s to early 1960s (McCullough, 1991), and CAF was developing in 1970s (Sato et al., 1987a). It is believed that ruminal pH does not fluctuate dramatically when cows fed with TMR. TMR provides a normal fermentation in rumen and thus decreases the incidences of metabolic disorders. When fed by CAF, cows proceed a steady digestion in rumen due to frequently feeding. The ruminal pH fluctuation thus decreases (Sato et al., 1987b). The two feeding systems, therefore, can improve health and milk yield of cows (Baxter et al., 1971; Nocek and Braund, 1985; Sato et al., 1987b). Nevertheless, the two systems have their own limitations. When feeding with TMR, for instance, lactating cows have to be grouped basing on their milk yields in order that the cows may consume as much feed as they deserve. To meet the requirement of nutrients for cows, the diets should be adjusted frequently in coupling with the variation of nutrient content in feedstuffs and nutrient requirement or intake for cows at different physiological and lactation stages (McCullough, 1991). One of the main limitations of CAF is that the social dominants in a herd may repel the inferior ones, and steal the ration away from the latter (Sato et al., 1987a). As a result, the inferior ones cannot obtain as much nutrients as they require.

In warm seasons, cows prefer ingesting concentrate (Hsu and Lee, 1995) and tend to decrease feed intake due to heat stress (Gengler et al., 1970). When fed with TMR, cows might have no chance to adjust the intake ratio of roughage to concentrate by themselves. Therefore, the diets prepared for lactating cows should be frequently adjusted in coupling with the fluctuation of feed intake so that deficiency or imbalance of nutrients intake would not occur. Although CAF leaves the room for cows to adjust the roughage: concentrate ratio by themselves, roughage intake is likely to be insufficient because cows prefer concentrate rather than roughage. It may thus cause acidosis and which in turn affects the health of the cows.

The thermoneutral zone for Holstein cows is from 5 to 25°C (NRC, 1981). High relative humidity jeopardizes cows even more in ways of physiology and production. Factors such as high ambient temperature, increase of heat increment (HI) by feeding more roughage, and increase of physiological activities for more milk production altogether make cows suffering from high milk yield in more extent (Johnson and Vanjonack, 1976). Climatically, Taiwan is not an ideal place for dairy cow farming because of its geographical location in the subtropical area. It is therefore quite necessary to find out a more appropriate feeding

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regime, which might be quite different from the ones for temperate area, to improve milk production of dairy cows in Taiwan under warm season.

MATERIALS AND METHOD

Feeding trial

Twenty-one lactating Holstein cows, 9 primariparities and 12 multiparities, were randomly allotted into three feeding regimes in a Completely Randomized Design according to their milk yield, parities, and calving time. The three feeding regimes were roughage fed *ad libitum* and concentrate fed either twice daily (2C), or four times daily (4C) as well as a total mixed ration (TMR) as shown in the following:

2C: Roughage ad libitum and concentrate fed twice daily.
4C: Roughage ad libitum and concentrate fed four times daily.
TMR: Fed *ad libitum* and which was comprised of 50% roughage and 50% concentrate with the same components and compositions as those in 2C and 4C.

The basal diet was prepared according to NRC (1989) plus 10% nutrient requirement as a safety allowance. The basal diet was comprised of two proportions: roughage and concentrate (table 1). The concentrate was made into 3mm pellets at 70°C after mixing weekly. Roughage and TMR were prepared at 8:00-9:00 daily. The moisture of the feedstuffs was monitored weekly for formulating the concentrate and the roughage.

The 21 cows selected a week prior to the experiment were 73±28 days in milk and at least 20 kg of milk yield. The average of the body weights and the 3.5% FCM yield of the cows were 555 kg and 25 kg, respectively. The cows were kept in individual stalls. The roughage of 2C, and of 4C as well as TMR was offered three times daily at 08:30, 16:30 and 22:00. The amount of the rations offered was parallel to the amount of intake keeping 2 to 3 kg leftover all time. The amount of concentrate fed in 2C and 4C was 1 kg per 3 kg of milk yield. For 2C, the concentrate was fed at 10:00 and 22:00 daily. For 4C, the concentrate was fed at 04:30, 10:00, 16:00, and 22:00 daily. Water was provided in bowl-shaped containers and freely accessed all time. The cows were milked twice daily at 04:30 and 16:00. The cows were released to an 800 m^2 pasture with shelters for resting or exercising during 04:00-07:30 and 13:30-16:30 everyday. During the experimental periods, the cows were dewormed once every half-month.

Two kilograms of feed samples were collected weekly and placed in a paper bag. They were dried at 60°C for 72 h and then stabilized moisture content in the air for 48 h. The samples thus prepared were so-called air-dried. The air-dried samples were ground by a grinder with 2 mm mesh and stored at -18°C for further analyses. The dry matter percentage (DM%) of the experimental rations was determined by heating at 110°C. The crude protein (CP%) was measured according to the AOAC method (1984). The measurements of neutral detergent fiber (NDF) and acid detergent fiber (ADF) were according to that by Van Soest et al. (1991). Dry matter intake (DMI) was determined of the total feed consumption multiplied by its dry matter content percentage. Milk samples were collected on one day every week during the experimental period. When sampling, 25 ml milk from each cow was collected on the morning and in the afternoon, and all the milk from each cow was well mixed. The milk composition was measured using the infrared Foss Electric Co. Milk Scan 255 A/B Type.

Body weights of the cows were measured once every four weeks. The body weight of each cow was determined by taking the average of the body weights measured at 08:00 on two consecutive days.

Data collected were analyzed with the General Linear Model Procedure of SAS (1985). Each cow was regarded as an experimental unit. The differences among the means were tested using Ducan's Multiple Range Test.

Ruminal trial

In a 3×3 Latin Square Design, three rumen fistulated

Table 1. Ingredient and composition of the basal diet[1]

Ingredients	Roughage	Concentrates
Corn silage	800	-
Alfalfa hay	200	-
Yellow corn	-	622
Soybean meal, 44% CP	-	232
Fish meal, 65% CP	-	70
Lard	-	30
Molasses	-	30
Limestone	-	6
Dicalcium phosphate	-	4
Salt	-	4
Premix[2]	-	2
Total	1,000	1,000
Nutrient calculated		
CP	127	203
NEL, Mcal/kg	1.3	2.1
Undegradable protein, g/kg protein	304	460
Nutrient analyzed		
CP	125	206
ADF	313	49
NDF	424	244

[1] On dry matter basis expressed as g/kg except of net energy.
[2] Each kg contains: Vit. A, 10,000 KIU; Vit. E, 70 KIU; Vit. D3, 1,600 KIU; Fe, 50 g; Zn, 40 g; Mn, 40 g; I, 0.5 g; Co, 0.1 g; Cu, 10 g; Se, 0.1 g.

Holstein dry cows were randomly allotted into the three feeding regimes as those in Feeding Trial. Each experimental period for treatment was 14 days, of which the first 11 days were adapting period and the 12th and 14th days were the sample-collecting dates. The three feeding regimes were applied to all over the three dry cows and to all over the three experimental periods.

Both the feeding trial and the ruminal trial were conducted during a hot season beginning on May 10 and ending on July 12. The average ambient temperature and its standard deviation was 29.94±2.21°C with a range from 22.8 to 33.1°C. The relative humidity was 83.1±6.7% with a range from 69.6 to 97.5%.

The average body weight of the three-ruminally fistulated cows was 600 kg. They were confined in a 50 m^2 cement-floored pen with fixing stalls. The amounts fed to the cows in 2C and 4C were 9.5 kg. Water was freely accessible. The ruminal environment was considered to be in a stable status after 11 days of adaptation to the feeding regimes. On the 12th and the 14th days, the ruminal fluid samples were collected right before feeding on the mornings and at 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 and 22 h post feeding. At sampling, about 200 mL ruminal fluid was collected via fistula from the bottom of the rumen. The ruminal fluid pH was measured with a Suntex TS-1 digital pH/mV meter immediately after the sample was collected. The ruminal fluid was then filtered with 4-layer cheesecloth and well mixed with 25% orthophosphate in 5:1 v/v ratio. The samples thus prepared were stored at -18°C for further analyses.

Ammonia content in the ruminal fluid was measured according to the method of Chaney and Marbach (1962). The measurement of individual VFA content in the ruminal fluid samples was based on the method by Erwin et al. (1961). The content of total VFA was calculated by taking the sum of all individual VFA including C2, C3, iso-C4, C4, iso-C5, and C5.

Data were analyzed with the General Linear Model Procedure of SAS (1985). The samples of a variable collected from each cow in each period were regarded as in the same experimental unit. The differences among the means of the treatments were tested using Ducan's Multiple Range Test.

RESULTS AND DISCUSSION

Feeding trial

For all the three feeding regimes, there were no significant differences (p>0.05) in roughage dry matter intake, concentrate dry matter intake, total dry matter intake (the sum of the former two intakes), ratio of roughage to concentrate, crude protein intake, and net energy intake.

The crude protein and the net energy intakes were not as much the amounts of 2.671 kg/d and 27.19 Mcal/d as recommended respectively by the NRC (1989). The reason why the intakes decreased was considered due to the effect of heat stress in the warm season (Gengler et al., 1970). The decrease in dry matter intake caused the coupling decreases in both crude protein intake and net energy intake. The decreases of these intakes consequently resulted in the decrease of milk yield (Giesecke et al., 1994).

Both the milk yield and 4% FCM was significantly higher in 4C than those in 2C and TMR (table 2). The result was possibly, at least in partial, due to that increase of concentrate feeding frequency to the cows could increase the digesting rate and which in turn provide more nutrients from time to time to sustain milk yield. Or, there was another possibility that the cows in 4C produced more milk at the expense of negative body weight gain even though there were no significantly different in body weight changes among the three feeding regimes shown in table 2. There were two factors resulted that the cows produced less milk than they did during pre-experiment. One was that the crude protein and the net energy intakes both did not meet the respective amounts of 2.67 kg/d and 27.2 Mcal/d as recommended by the NRC (Giesecke et al., 1994). The other one was that these cows were at their post peak stage of lactation when the milk yield was going down physiologically. If it was caused by the first factor, then the roughage dry matter intake should increase to meet the nutrient requirement for achieving the milk production potency when the feeding amount of concentrates was limited for the cows in 2C and 4C (Campling and Murdoch, 1966). However, the roughage dry matter intake did not increase as a result in 2C and 4C. This was probably due to the heat stress that suppressed the feed intake of the cows in warm season (Gengler et al., 1970).

The results showed that the three feeding regimes had no significant effects (p>0.05) on the milk compositions in terms of total solid, fat, protein and lactose percentages nor on milk fat, protein and lactose yields (table 2).

The effects of concentrate feeding frequencies and TMR on milk production efficiency were shown in table 2. The milk production efficiencies of 2C and 4C were significantly better than that of TMR. The reason was considered to be that total dry matter, net energy, and crude protein intakes in 2C and 4C all were relatively lower than those in TMR. Besides, milk and 4% FCM yields in 2C and 4C both were similar to or greater than those in TMR.

Table 2 showed the effects of the three feeding regimes on the body weight changes of the lactating cows. The average body weights of the cows in the three feeding regimes were losing during the first 4 weeks of the experimental period. However, the differences of the body weight loss among the three feeding regimes were not significant. During the late 4 weeks of the experimental

Table 2. Effect of concentrate feeding frequency of twice or four times daily versus total mixed ration on performance in lactating dairy cows (Feeding Trial)

Items	2C	4C	TMR	SEM	P value
Cows, head	7	7	7		
Initial body weight, kg	536.1	549.1	580.8	27.82	0.680
Body weight changes			(%)		
0-4 week	98.43	97.39	96.50	1.09	0.461
4-8 week	101.08	100.14	104.04	1.91	0.359
0-8 week	99.46	97.53	100.28	1.64	0.506
Feed intake and milk yield			(kg/d)		
Roughage DM intake[1]	6.08	6.72	6.64	0.08	0.101
Concentrate DM intake[2]	5.71	6.23	6.64	0.06	0.094
Total DM intake	11.79	12.83	13.28	0.39	0.065
R/C DM intake	1.11	1.12	1.00	0.03	0.099
CP intake	1.94	2.10	2.20	0.07	0.062
NE intake, Mcal/d	19.90	21.58	22.58	0.75	0.062
Milk yield	17.31^b	18.84^a	17.59^b	0.01	0.029
4%FCM	15.67^b	17.12^a	15.64^b	0.33	0.041
Milk composition			(g/kg)		
Total solid	120.3	121.0	119.9	0.23	0.919
Fat	34.0	34.1	32.7	0.01	0.753
Protein	32.0	31.8	32.8	0.04	0.211
Lactose	47.4	46.3	47.4	0.07	0.231
Milk composition yield			(kg/d)		
Total solid	2.08	2.25	2.11	0.08	0.084
Fat	0.58	0.64	0.57	0.02	0.127
Protein	0.55	0.60	0.57	0.02	0.131
Lactose	0.82	0.87	0.83	0.03	0.301
Milk production efficiency					
DM intake/milk yield	0.69^b	0.69^b	0.77^a	0.03	0.003
DM intake/4%FCM	0.76^b	0.76^b	0.87^a	0.03	0.0001
NE intake/4%FCM	1.29^b	1.28^b	1.49^a	0.05	0.001
CP intake/4%FCM	0.13^b	0.12^b	0.14^a	0.005	0.001
CP intake/milk protein yield	3.56^b	3.55^b	3.89^a	0.15	0.039

^{a,b} Means in the same row without the same superscripts significantly differed ($p<0.05$).
[1] Roughage dry matter content was 34.8%.
[2] Concentrate dry matter content was 90.3%.

period, the cows were gaining their body weights. However, the differences of the average body weight gains among the three feeding regimes were again not significant. During the whole experimental period of 8 weeks, there were no significant differences among the three feeding regimes in body weight changes. For each 1kg of body weight loss, it is equivalent to 7 Mcal net energy (Moe et al., 1970). If the net energy is mainly used to synthesis of milk, the effect of rations on milk production will be distorted. To detect the extent of the possible effect of milk production sustained by the net energy coming from body weight loss, MANOVA analysis was included in the GLM procedure in this experiment. However, the result indicated that there was no significant partial correlation between the body weight changes and the milk or FCM yields.

Ruminal trial

The effects of feeding regimes on ruminal fluid pH, VFA and ammonia nitrogen concentrations were shown in table 3. The fluctuation of the ruminal pH along with time arouse by feeding was shown in figure 1. The ruminal pH in TMR was in a range as narrow as from 6.1 to 6.6, which was fluctuating in a range not so much as those in 2C and 4C. The ruminal pH of the cows in 2C at 4 h after feeding with concentrate declined to the lowest of 5.85. The pH in 4C decreased to the lowest of 5.95. The averages of the ruminal pH in the three feeding regimes were close to each other ($p>0.05$) (table 3). Church (1979) indicated that ruminal pH would be as low as 5.20-5.45 during 2-6 h after feeding. The result of this experiment was in agreement with that by Church (1979) in that it took the same time (2-6 h) post feeding to reach the lowest rumianl pH. The readily fermenting carbohydrates in concentrate such as sugar and starch degrading into organic acids is the cause of lower ruminal pH (Church, 1979). The lowest rumianl pH in this experiment was a little greater than 5.20-5.45 reported by Church (1979). It was at least in part, if not entirely, due to the less dry matter intake of the concentrates in this experiment. Cows that consume more feeds in either concentrate or roughage would have greater fluctuation in ruminal pH (Rumsey et al., 1970).

Before feeding, ruminal pH in TMR was significantly higher than that in 4C ($p<0.05$). At 4 h post feeding, ruminal pH in TMR was higher than that in 2C ($p<0.05$). However, the differences of the average daily ruminal fluid pH among the three feeding regimes were not different ($p>0.05$) (table 3). The ruminal fluid pH diurnal fluctuation base lines of TMR, 4C and 2C kept above 6.1, 6.0 and 5.9, respectively. Besides for each feeding regime, the time interval that the ruminal fluid pH keeping close to the lowest level was no longer than 4 h.

The changes in ammonia nitrogen concentrations of ruminal fluid were shown in figure 1. The diurnal ammonia-N concentration of TMR varied from 18.4 to 27.6 mg/dL. The less fluctuating of the diurnal ammonia-N concentrations in TMR and 4C might be due to the more frequent intake although they were not statistically different. The result was in consistent with that by Sato et al. (1987b).

Two hours post concentrate feeding, the ammonia-N concentrations in the ruminal fluid from the cows of 2C stroke a peak and gradually declined thereafter. The ruminal fluid ammonia-N concentration of 2C was higher than that of 4C ($p<0.05$) at the same time. Bunting et al. (1987) stated that ruminal fluid ammonia-N concentration decreased as the concentrate feeding frequency increased.

The ammonia-N concentration in 4C varied from 10.3 to 13.6 mg/dL in a day. The variation range was the least one among the three feeding regimes. One possible explanation for the declined variation of ammonia nitrogen concentration was that the feeding frequency of concentrates increased (Bunting et al., 1987). Another explanation was that it might be due to the less dry matter intake from concentrates as reported by Bartley et al. (1976).

Although the ammonia nitrogen concentration in cows feeding to TMR was the lowest among the three feeding regimes, it was residing within the normal range as 0.39-29 mg/dL (Satter and Slyter, 1974). The heavily variation of ammonia nitrogen concentration in TMR was probably due to intake from time to time. Cecava et al. (1990) indicated that ruminal ammonia concentration maintained at a very narrow range such as 17.2-18.3 mg-N/dL in steers fed 12 times daily comparing to the great variation ones such as 37.4, 30.5, 14.7, 13.3 and 14.6 mg-N/dL at 1, 3, 6, 9 and 11 h, respectively, after feeding when the animals fed twice daily. It is, therefore, concluded that the less the feeding frequency is the greater the fluctuation in amonia nitrogen concentration.

Change in total VFA concentration

The total VFA concentrations before and after feeding in ruminally fistulated cows fed with 2C, 4C, and TMR were shown in figure 1. The diurnal total VFA concentrations in 2C and 4C vibrated up and down. The daily change in VFA concentration was due to irregular feeding time. At each individual time slot, 2C's VFA concentration was higher than 4C by 6 hours after feeding ($p<0.05$). Bath and Rock (1963) indicated that when cows were fed once daily, their VFA concentrations reached maximum at 2 h after feeding.

Table 3. Effect of concentrate feeding frequencies of twice or four times daily versus total mixed rations on rumen fluid pH, ammonia nitrogen level, and volatile fatty acid composition (Ruminal trial)

Ruminal fluid characteristics	Feeding regime			SEM	P value
	2C	4C	TMR		
pH	6.1	6.1	6.2	0.17	0.641
Ammonia-N (mg/dL)	14.4	11.7	10.9	3.97	0.186
Total VFA concentration (mmole/L)	111	110	96	11.95	0.663
VFA composition (Molar%)					
Acetic acid	62.2	58.0	65.8	4.80	0.434
Propionic acid	20.0	26.1	20.0	4.69	0.399
Butyric acid	13.0	11.7	10.3	0.72	0.897
Acetic acid : propionic acid	3.17	2.75	3.49	0.66	0.388

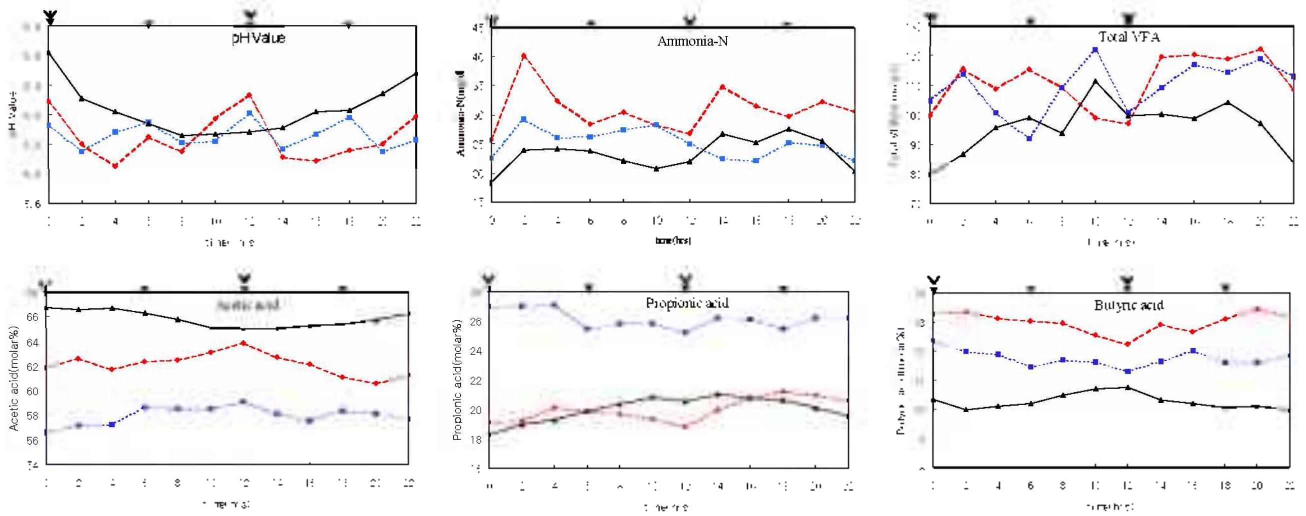

Figure 1. Effects of concentrate feeding frequency of twice or four times daily versus total mixed ration on ruminal pH value, ammonia nitrogen concentration, total VFA concentration, as well as the molar percentage of acetate, propionate and butyric acid. ▼ and ↓ express feeding time in the 4C and 2C groups, respectively. –◆–, ···■···, –▲– represent 2C, 4C and TMR feeding regimes, respectively.

and maintained 4-6 h before they started decreasing.

The averages of total VFA concentrations in 2C, 4C, and TMR were shown in table 3. They were not significant different. According to French et al. (1990), when six Holstein cows were fed with concentrate two times daily, twelve times daily, and fed with TMR, their total VFA concentrations were 87.3, 79.2 and 71.0 mmole/L respectively.

Change in individual VFA

The variations of ruminal fluid's C_2, C_3, and C_4 molar % measured before and after feeding for ruminally fistulated cows fed with 2C, 4C, and TMR were shown in figure 1. Acetic: propionic acid ratio was shown in table 3. TMR's C_2 molar % was higher than 4C's before feeding ($p<0.05$, figure 1). Although TMR's C_2 molar % was higher than 2C's at any time and 2C's was higher than 4C's, the differences among the three feeding regimes were not significant ($p>0.05$). The daily average C_2 molar% for 2C, 4C and TMR were obviously close to each other (table 3).

Propoonic acid molar % in 4C was higher than theose in the other two feeding regimes at any time; however, there was no significant difference ($p>0.05$, figure 1). The average daily propionic acid molar % for 2C, 4C, and TMR were not significantly different (table 3).

Butyric acid molar % in 2C was higher than that in TMR before feeding and at 6, 8, 10, 14, 16, 18, 20 and 22 h after feeding ($p<0.05$, figure 1). Although 2C's figure of C_4 molar % was higher than 4C's and 4C's were higher than TMR's, their differences were not significant ($p>0.05$). The average daily C_4 molar % for 2C, 4C and TMR were not significantly different (table 3).

Bath and Rook (1963) indicated that when increasing frequency of feeding, the VFA concentration of castrated cows displayed a temporary climax. However, the individual VFA compositions in ruminal fluid were not affected significantly by the three feeding regimes.

CONCLUSION

In this experiment, milk yield and 4% FCM in 4C were noticeably higher than those in 2C and TMR ($p<0.05$). Milk production efficiency in 4C was better than those in 2C and TMR. These results implied that for the best milk yield concentrate fed four times daily and roughage fed *ad libitum* were the best feeding regime for lactating cows which had medium milk yield in hot season.

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