Dietary Supplementation of Fat Increased Milk Fat Percentage without Affecting Ruminal Characteristics in Holstein Cows in a Warm Tropical Environment

A. S. Wang1,2, D. F. Jan1, K. J. Chen1,4, D. W. Yang2 and Y. K. Fan1,*

1Department of Animal Science, National Chung Hsing University, Taichung 402, Taiwan, ROC

ABSTRACT: The purpose of this experiment was to investigate the effect of the diets supplemented with lard or prilled fat (Carolac®) on lactation performance, plasma constituents and ruminal characteristics of Holstein cows under a warm climate. In trial 1, 18 Holstein cows, 14 primiparas at 43 DIM and 4 multiparas at 55 DIM, were randomly assigned into six 3 × 3 Latin squares, in which three dietary treatments were isoproteinous but varying in energy contents and three 21 d periods. The treatments were basal diet (Control), basal diet supplemented with 2.5% lard (LD), and basal diet supplemented with 2.5% commercial Prilled fat (PF). In trial 2, three rumen canulated pregnant nonlactating Holstein cows with 550 kg average body weight were allotted into a 3 × 3 Latin square design with diets same as in trial 1 were fed to the cows at the level of 1.5% body weight on dry matter (DM) basis. The results indicated that the DM intake did not differ among the treatments. Milk yield and 4% FCM yield were greater (p<0.05) in PF than in Control. LD and PF resulted in greater milk fat percentage. Protein, lactose and solid contents in milk were not different among the three dietary treatments. The concentration of nonesterified fatty acids (NEFA) in plasma was significantly greater in LD and PF than that in Control. However, the concentrations of triglycerides, urea nitrogen, and cholesterol in plasma were not significantly different among the three treatments. Although the ruminal molar percentage of isobutyrate in LD and PF was greater, no significant difference was observed in ruminal pH, NH3-N concentration and VFA production among the three treatments. Diet supplemented with fat can improve milk yield and milk fat percentage without resulting in disadvantages of ruminal characteristics in cows at early lactation and under warm climate.

Key Words: Holstein Cows, Dietary Fat Supplementation, Milk Production, Ruminal Characteristics

INTRODUCTION

As dairy herds are being improved genetically, the productive potential of milk yield is keeping dramatically increased. The high production dairy cows are inevitably in a state of negative energy balance during early lactation. It is especially true that dairy cows cannot sufficiently fulfill their high production potential in tropical and subtropical areas since heat stress decreases their feed intake during warm seasons. In these areas, energy is usually the major limiting factor in milk production of lactating dairy cows, thus, a strategy usually adopted is increase of dietary concentrate ratio to meet the energy requirements for lactation. However, overfeeding of concentrate could result in acidosis and lowering the ruminal pH, which in turn is deleterious to the microbial growth and fiber digestion in rumen. These effects consequently result in low milk fat (Elliott et al., 1995).

* Corresponding Author: Y. K. Fan. Tel: +886-4-2285-3748, Fax: +886-4-2286-0265, E-mail: ykfan@dragon.nchu.edu.tw
2 Livestock Research Institute, Council of Agriculture, Executive Yuan, Taichung 712, Taiwan
3 Central Taiwan Office, Council of Agriculture, Executive Yuan, 8, Kuang Hwa Road, Chung Hsing Hsing Tsuen, Nantou 540, Taiwan.
4 Taitung Breeding Animal Propagation Station, Taiwan Livestock Research Institute, Council of Agriculture, 18, Bing Lang Tsuen, Peinan, Taitung 954, Taiwan.

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Most of the researches pertaining to lipid supplementation to diet for lactating dairy cows were conducted in temperate area. When applying the technologies established by temperate area to tropical or subtropical area, the results not necessarily occur as what they are expected. Our previous study provided an example of this out-of-expectation (Fan et al., 2002). Although a lot of similar researches have been done in temperate areas, researches are still needed to meet the specific demands and to fit the unique environment of the local areas in tropical or subtropical.

Fat contains much more energy than that of grains for feeding cows. Andrew et al. (1990) indicated that one commercial fat, Ca-Soap, made of palm fatty acid, contained 85% fatty acids, which provided 3.3 times of NEL as did corn on the dry basis. At the equivalent level of DM intake, ration supplemented with fat results in more energy intake. Thus, supplementation of fat in the diet for early lactating cows may improve their negative energy balance, prevent them from body weight loss and increase their milk yield (NRC, 1989; Palmquist and Jenkins, 1980).

According to physical and chemical properties, fat can be classified into three categories, namely, plant oils that are highly unsaturated, animal fats that are highly saturated and protected fats that are for commercial use. Overuse of unsaturated fatty acids in diets could cause disturbance to the ecology of ruminal microbes and their fermenting activities, which would further result in decline of milk...
production efficiency (Coppock and Wilks, 1991). To prevent these disadvantageous effects, some general key ways should be adopted such as using protected fat, say, prilled fat or using unprotected fat in oilseeds or tallow with limitations of no more than 4 to 5% of DM or no more than 7 to 8% of total crude fat (Coppock and Wilks, 1991; Palmquist and Conrad, 1978).

Acetate:propionate in rumen fluid of midlactation cows obtained by an esophageal tube decreased with increasing dietary prilled fatty acids containing 0, 3, 6 or 9% of dry matter as reported by Ferguson et al. (1990). Addition 3 to 5% fat to common feeds appears to be tolerated by ruminal microorganisms (Palmquist, 1984). Similarly, Chalupa et al. (1986) recommended that 6 to 8% supplemental long-chain fatty acid was probably sufficient to optimize productivity and health of Holstein cows since they found that addition of fatty acids at 10% to a basal diet resulted in dramatic decrease of acetate to propionate ratio by 50 to 60%. These researches show that lipid supplementation in the diet above certain level may affect ruminal fermentation. Thus, a proper level in addition lipid to the diet that would not cause adverse effects on ruminal fermentation is an issue of interest for this study.

Tallow contains about 45% saturated fatty acids. Lard contains 40% saturated fatty acids, which is similar to tallow in physical and chemical properties. However, lard is scarcely supplemented in diets for dairy cattle in practice. The production of lard is much more abundant than that of tallow in Taiwan because of a large national swine industry. Lard might be a good source of energy for dairy cattle. However, there have been only a limited number of studies examining the use of lard in dairy cattle diets. This study was conducted to investigate the comparative effects of dietary supplementation of lard (unprotected fat), or prilled fat (a commercial protected fat) on lactational performance, especially focusing on milk and milk fat yields as well as comparative changes in blood and ruminal parameters.

**MATERIAL AND METHODS**

**Feeding trial**

Eighteen Holstein cows, 14 primiparas at 43 DIM and 4 multiparas at 55 DIM, were randomly allotted into 3 dietary treatments in six 3×3 Latin squares. A basal diet (control) was formulated according to NRC (1989) plus 10% nutrient requirement as a safety allowance basing on nutrient requirement for lactating cows with 490 kg average body weight and 25 kg daily milk yield. LD represented the basal diet supplemented with 2.5% lard to replace equal amount of yellow corn. PF represented the basal diet supplemented with 2.5% prilled fat, a commercial fat, to replace equal amount of yellow corn. The three treatment diets were, therefore, isoprotenous but varying in energy contents as shown in Table 1. Each experimental period was 21 d including 14 d for adaptation and 7 d for data collection.

The treatment diets were fed in form of total mixed ration (TMR) and were prepared daily at 8:00 am. The concentrate: roughage ratio of the TMR was 50:50. The TMRs were provided three times per day at 9:00, 17:00 and 21:00 h, respectively. The TMRs were fed ad libitum and individually so as to maintain 2 to 3 kg leftover in the feed troughs always. The feed consumption was calculated as amount provided subtracted by amount of leftover. Drinking water provided with water bowls was freely accessible.

The cows were placed in individual stanchions in a house equipped with two big electrical fans to decrease the extent of heat stress. The daily variations in ambient temperature and humidity were recorded with an automatic recorder (Sato Sigma II Model NS II-Q). During the experimental period, the averages of ambient temperature and humidity were 27.5±3.4°C and 70.6±15.6%,
respectively. The cows were released to an 800 m² earth-floored lot with shelters for resting or taking exercise during 5:30 to 9:00 h and 13:00 to 16:00 h daily. Deworming external parasites was applied to the cows once for half a month.

Two kg of each of the three experimental diets was sampled weekly. The air-dried samples were obtained by being placed in paper bags and dried in a 60°C windy forced oven for 72 h followed by sitting at ambient temperature for 48 h for constant moisture content. The air-dried samples were ground with a mill of 1 mm screen. The ground air-dried samples were stored at -18°C for further analyses. The determination of crude protein (CP%) followed the method of AOAC (1984). The determinations of neutral detergent fiber (NDF) and acid detergent fiber (ADF) followed the methods of Van Soest et al. (1991).

The cows were milked twice daily at 5:00 and 16:00 h. The milk yield was measured with a computerized automatic device (GM 2000) during each time of milking. Milk sample of 25 ml for each milking was collected during the last 3 d in each experimental period. The milk samples from each cow in each experimental period were intermingled and preservatives added for storage. The contents of milk compositions, namely, fat, protein, lactose and total solids, were determined with Milk Scan (Milk Scan 255 A/B Type, Foss Electric Co.).

Blood sample was collected every other day during the last 5 d in each experimental period according to Roseler et al. (1993). A heparinized vacutainer was used for collecting blood sample via tail vein at 3 h post feeding on the morning of collection day. The blood plasma was obtained by centrifugation at 900×g for 10 min. The plasma was stored at -20°C for further determinations of free fatty acids, triglycerides, total cholesterol and urea-N.

Aliquots of 100 µL blood plasma, 3 ml extraction solvent, which was comprised 49% (vol) chloroform, 49% (vol) heptane and 2% (vol) methanol, and 1 ml copper reagent, which was comprised 2.42 g Cu(NO₃)₂·3H₂O and 5 ml triethanolamine diluted with saturated NaCl solution for 100 ml and pH 8.3, were mixed in a test tube with cover. The mixed liquid was proceeding twice of shaking for 2 min and sitting for 1 min. Supernatant was obtained by centrifugation the mixed liquid at 900×g for 5 min. Mixing 1 ml of the supernatant with 0.25 ml color reagent, 10 mg tan dissolved in ethanol for 100 ml, and then let the solution sitting for 5 min. The absorbance of the solution was determined with a spectrophotometer at 570 nm. The concentration of free fatty acids in plasma was determined by comparing the absorbance with that of standard solution containing palmitic acid.

Triglycerides in blood plasma were determined spectrophotometrically with a kit (Roche Co., Sweden) at 500 nm. Total cholesterol and urea nitrogen (PUN) contents in blood plasma were determined with an Automatic Analyzer (Hitachi 7050, Japan) plus kits from Roche Co. (Sweden) and National Co. (Japan), respectively.

Ruminal trial

Each of the three rumen canulated Holstein dry cows with average body weight of 550 kg was individually kept in a 25 m² concrete-floored pen. Each pen had a crate for fixation purpose. The experimental rations and the experimental period were the same as in feeding trial. The experimental rations were prepared in form of TMR and were provided with the level of 1.5% body weight on the DM basis. The daily amount of the TMR was split into two equal parts and given at 9:00 and 21:00 h. Drunking water was freely accessible.

Ruminal juice of 250 ml was collected into a 500 ml flask via bottom of the ventral sac through canula on the morning right before feeding (0 h), and 3, 6, 9 and 12 h post feeding daily for the last 3 d in each experimental period. The pH of the collected ruminal juice was immediately measured using a pH meter (WPA Linton Cambridge Model CD720) followed by filtering through 4 layers of gauze. For reaching pH 2, 50 ml of the filtered ruminal juice was mixed with 1 ml 50% H₂SO₄ and the aliquots of the mixture were stored at -20°C for further determinations. The ammonia-N was determined according to the method of AOAC (1984). The determinations of VFAs followed the method by Erwin et al. (1961). Briefly, the thawed ruminal juice aliquot 6 ml was centrifuged at 10°C and 5,000 rpm for 20 min. The aliquot of the supernatant 6 µL was injected in a gas chromatographer (Hitachi G-5000A). The conditions used were 125°C at the column in the oven, 180°C at the injector and 200°C at the detector as well as using He as carrier gas with pressure at 5 kg/cm² and flow rate at 40 ml/min, H₂ with pressure at 3 kg/cm² and flow rate at 23 ml/min, and air with pressure at 4 kg /cm² and flow rate at 450 ml/cm². The integrator used was Hitachi D-2,500 with the conditions of Plot ATT2 and chart speed at 0.5 cm/min. The including of the column was 15% SP-1200 with 1% H₃PO₃. The peaks of VFAs in ruminal juice were compared with those in the standard solution (Spelco WSFA-2) to calculate their contents (mmole/L) and their molar percentage individually.

Statistical analysis

Both in feeding and ruminal trials, the data were statistically analyzed using General Linear Model Procedure (GLM) of SAS (1989). Each experimental period in each cow was regarded as an experimental unit. During the experimental period, if the cows were in abnormal status, such as in mastitis, the data collected would be regarded as missing data. The significances of the differences among the means of the treatments were tested by Least Square Means procedure.
RESULTS AND DISCUSSION

Feeding trial

Dry matter intakes were not significantly different whether the rations supplemented with fat or not (Table 2). Pantoja et al. (1994) indicated that dry matter intake was linearly decreasing along with the increases of dietary supplementation of unsaturated fatty acids in lactating cows. The result might be attributable to the effect of coating on fibers by considerable amount of unsaturated fatty acids, which are consequently and adversely influencing ruminal fermentation and passage rates. Schauff and Clark (1992) also found the similar results when cows were fed with rations containing 3, 6 or 9% of protected fat, say calcium long chain fatty acids, and attributed the decrease of feed intake to the worse palatability of the supplemented fat. However, rations supplemented with fat would not necessarily cause the decrease of dry matter intake during early lactation (Wu et al., 1993). These researches implied that the effect of fat supplementation in diets on dry matter intake was depending on the forms, extent of saturation and amount of fat supplemented. In this study, the cows were at 46 DIM, belonging to the stage of early lactation and the amount of fat supplemented was 2.5%, a low level of supplementation. Under these conditions, the dry matter intake was not adversely affected by the supplementation of low level and more saturated fat such as lard or Prilled fat during early lactation.

Cows fed PF produced more milk (7%) and milk fat (15.8%) than those on the control ration, with cows consuming LD intermediate and not different from either the control or PF ration. Similar results were observed with FCM, with PF producing 12.1% more FCM compared with the control diet. Diet supplemented with PF improved (p<0.05) efficiency of milk production, expressed as DM intake/kg FCM yield. Diet supplemented with 3 to 4% fat results in increase of milk yield 2 to 12% (Casper et al., 1990; Coppock and Wilks, 1991).

The records in this experiment showed that the averages of daily highest ambient temperature and humidity were over 30.9°C and 86.2%, respectively during the experimental period. Converting these figures into temperature-humidity index (THI), it was 85, an index of intermediate stressful status. The milk yield during experimental period (warm season) was depressed in comparison with that in cool seasons (Ting, 1990) also serving as a side-evidence to indicate that the cows were being in the status of heat stress. Cows consuming PF produced more milk, however, consumed a similar intake with no differences in body weight change compared with those consuming control diet.

The results showed that supplementation with PF increased energy for lactation. Grummer and Carroll (1991) suggested that increase energy density of rations for lactating cows by supplementation of fat improved energy balance, and which consequently resulted in more energy available to support milk production of genetic potentials. Cant et al. (1993) discussed more details about the beneficial merits of supplementing genetic potentials.

Table 2. Effect of dietary supplementation of fat on dry matter intake, milk yield and milk composition of lactating Holstein cows (Feeding trial)

<table>
<thead>
<tr>
<th>Dietary treatment</th>
<th>Control</th>
<th>LD</th>
<th>PF</th>
</tr>
</thead>
<tbody>
<tr>
<td>DMI kg/d</td>
<td>21.4±1.69</td>
<td>18.6±1.81</td>
<td>18.2±1.90</td>
</tr>
<tr>
<td>Milk kg/d</td>
<td>22.9±0.46&lt;sup&gt;b&lt;/sup&gt;</td>
<td>23.9±0.50&lt;sup&gt;b&lt;/sup&gt;</td>
<td>24.6±0.52&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>4% FCM, kg/d</td>
<td>20.6±0.53&lt;sup&gt;b&lt;/sup&gt;</td>
<td>21.9±0.56&lt;sup&gt;b&lt;/sup&gt;</td>
<td>23.1±0.59&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>DMI/milk, kg/kg</td>
<td>0.96±0.073&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.79±0.078&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.74±0.082&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>DMI/4% FCM, kg/kg</td>
<td>0.98±0.046&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.87±0.049&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.81±0.051&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>BW change, kg/d</td>
<td>-0.17±0.145</td>
<td>-0.15±0.147</td>
<td>0.17±0.153</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Milk composition</th>
<th></th>
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</tr>
</thead>
<tbody>
<tr>
<td>Fat, %</td>
<td>3.28±0.050&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.45±0.055&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.55±0.058&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Fat, kg/d</td>
<td>0.76±0.023&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.82±0.025&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.88±0.026&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>CP, %</td>
<td>2.86±0.028</td>
<td>2.86±0.029</td>
<td>2.81±0.031</td>
</tr>
<tr>
<td>CP, kg/d</td>
<td>0.66±0.015</td>
<td>0.68±0.016</td>
<td>0.69±0.017</td>
</tr>
<tr>
<td>Lactose, %</td>
<td>4.52±0.064</td>
<td>4.67±0.068</td>
<td>4.61±0.071</td>
</tr>
<tr>
<td>Lactose, kg/d</td>
<td>1.05±0.023</td>
<td>1.09±0.025</td>
<td>1.12±0.026</td>
</tr>
<tr>
<td>Solids, %</td>
<td>11.8±0.13</td>
<td>11.7±0.14</td>
<td>11.7±0.15</td>
</tr>
<tr>
<td>Solids, kg/d</td>
<td>2.73±0.069</td>
<td>2.78±0.073</td>
<td>2.86±0.077</td>
</tr>
</tbody>
</table>

Control: basal ration. LD: basal ration supplemented with 2.5% lard. PF: basal ration supplemented with 2.5% prilled fat.

Experimental unit: each experimental period in each cow was regarded as an experimental unit.

<sup>a,b</sup> Means (±SEM) within the same row without the same superscripts are significantly different (p<0.05).
Table 3. Effect of dietary supplementation of fat on concentrations of lipids and urea nitrogen in plasma of lactating Holstein cows (Feeding trial)

<table>
<thead>
<tr>
<th>Dietary treatment</th>
<th>Control</th>
<th>LD</th>
<th>PF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Experimental unit</td>
<td>18</td>
<td>17</td>
<td>17</td>
</tr>
<tr>
<td>NEFA</td>
<td>30.9±0.79&lt;sup&gt;b&lt;/sup&gt;</td>
<td>33.9±0.84&lt;sup&gt;a&lt;/sup&gt;</td>
<td>34.0±0.84&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Triglyceride</td>
<td>11.0±1.88</td>
<td>11.8±1.81</td>
<td>9.5±1.81</td>
</tr>
<tr>
<td>Cholesterol</td>
<td>101±4.5</td>
<td>106±4.8</td>
<td>103±4.8</td>
</tr>
<tr>
<td>PUN</td>
<td>14.8±0.39</td>
<td>14.9±0.41</td>
<td>14.6±0.41</td>
</tr>
<tr>
<td></td>
<td>mg/dl plasma</td>
<td>mg/dl plasma</td>
<td>mg/dl plasma</td>
</tr>
</tbody>
</table>

Control: basal ration. LD: basal ration supplemented with 2.5% lard. PF: basal ration supplemented with 2.5% prilled fat.
Experimental unit: each experimental period in each cow was regarded as an experimental unit.
NEFA: non-esterified fatty acids. PUN: plasma urea nitrogen.
<sup>a,b</sup>Means (±SEM) within the same row without the same superscripts are significantly different (p<0.05).

production and production efficiency also.

Comprehensively, ration supplemented with protected fat such as Prilled fat improve milk yield and milk production efficiency for cows in early lactation. Dietary supplementation of fat, either lard or Prilled fat, increased milk fat percentage. Prilled fat supplemented ration resulted in increase of milk fat yield. Cows consuming LD tended (p=0.08) to have increased milk fat yield. Similar results in relation to effect of dietary supplementation of fat have been reported elsewhere (Jenkins and Jenny, 1989; Schauf and Clark, 1992). Early reports indicated that diets supplemented with fats would decrease milk fat percentage. They attributed the adverse effect to large amount of polyunsaturated long-chain fatty acids appeared in rumen, by which the concentration of trans-isomers of these fatty acids increased during hydrogenation (Casper et al., 1988), which in turns inhibited the activity of acetyl-CoA carboxylase in mammary glands. Milk fat percentage was thus decreased due to the decline of resynthesis in short- and median-chain fatty acids catalyzed by the carboxylase (Palmquist and Jenkins, 1980). Comparing to content of about 80% unsaturated fatty acids in oils of plant sources, lard contains less unsaturated fatty acids (60%) accompanying with 40% saturated fatty acids. On the other hand, Prilled fat has been commercialized and is a protected fat, which may exert effects to prevent it from the modification by microbes and/or the adverse effect on microbial fermentation in rumen. The beneficial effects of promotions in milk fat percentage by dietary supplementation of lard or Prilled fat and milk fat yield by dietary supplementation of Prilled fat in this study may be attributable to the previously mentioned rationales.

Ration supplementation with LD or PF resulted in no significant differences in both percentage and yield of milk protein (Table 2). The results were consistent with reports by Drackley et al. (1992) and Firkins and Eastridge (1992). However, some researchers have reported that rations supplemented with fat would result in decrease of milk protein percentage by 0.10-0.15% (Casper et al., 1988; Canale et al., 1990; DePeters and Cant, 1992). Since the mechanism of how dietary supplementation of fat influencing milk protein percentage has not been exactly well elucidated (Canale et al., 1990; Karunananda et al., 1994), more efforts are needed to be put on this subject.

The lactose percentage, lactose yield, total solid percentage, and total solid yield in milk were not different either among the treatments. The results displayed that milk compositions and their respective yields, except of milk fat percentage and yield, were not altered by dietary fat supplementation.

Rations supplemented with lard or Prilled fat significantly increased plasma NEFA (Table 3), which were in consistent with the report by Grummer and Carroll (1991). Yang et al. (1978) indicated that NEFA were generated from body fat and dietary triacylglycerols undergoing hydrolysis through activation of lipoprotein lipase. Dietary supplementation of fat promotes the activity of lipoprotein lipase and increases insulin resistance as well as decreases esterification of fatty acids during triacylglycerols biosynthesis (Palmquist and Moser, 1981). These physiological responses consequently result in increases of blood NEFA contents, which might be providing sufficient precursors for milk fat synthesis in mammary glands.

Research by Grummer and Carroll (1991) suggested that fatty acids from dietary source require blood cholesterols for their transportation. Dietary supplementation of fat thus increases plasma concentration of cholesterol (Palmquist and Conrad, 1978; DePeters et al., 1989). However, no significant differences were observed among the three treatments in this study. One of the attributable rationales for the result was a short period being carried out in this study comparing to 15 to 18 wk were used by Smith et al. (1978) and Wrenn et al. (1978), who indicated that dietary supplementation of tallow or protected tallow increased concentration of plasma cholesterol level in lactating cows.

The optimal range of blood urea nitrogen concentration is 12 to 17 mg/dl, maximally no more than 20 mg/dl suggested by Stevensen (1994). Supplementation of fat in diet would not result in alteration of BUN (Drackley and Schingoethe, 1986; Kim et al., 1991; Maiga et al., 1995).
which was consistent with the result in this study. The three rations of the treatments were isoproteinous but varying in energy contents due to dietary fat supplementation. When the rations were fed to the lactating cows no significant alterations in PUN were observed and the variations of PUN were in a normal range. The results implied that the amount of fat supplemented in diets did not exerted substantial influence on the degradation and utilization of dietary proteins by ruminal microbes.

Ruminal trial

The ruminal pH, ammonia nitrogen and VFAs affected by dietary supplementation of fat are shown in Table 4. Dietary supplementation of fat exerted no substantial effect on ruminal pH.

Total VFA concentration was not significantly different among the three treatments (Table 4). The result implied that ruminal fermentation was not substantially influenced by dietary supplementation of fat. Since dietary fat serving as one of high-energy sources for feeding ruminants, its influences on ruminal fermentation should be well understood so that it could be utilized efficiently and beneficially. The effect of fats on ruminal fermentation is depending on their sort and amount used, particularly on their fatty acid composition, content and saturation of free fatty acids (Palmquist and Jenkins, 1980). Commercialized fats, such as Ca-LCFA and Prilled fat, which have been ruminally inactivated, have been effectively decreased, or even excluded, in their adverse effects on ruminal fermentations when diets are supplemented of these fats (Chalupa et al., 1986; Schauff and Clark, 1992). Palmquist (1984) indicated that tallow, containing about 50% of saturated fatty acids, comparing to free fatty acids exerted less adverse effects on ruminal fermentation. Although the content of saturated fatty acids in lard is close to that in tallow and Prilled fat is one of a protected fats, both of them did not influence the ruminal fermentation in this study giving previous statements the proofs.

The molar percentage of each VFA was shown in Table 4. No significant differences were found in molar percentages of acetate and propionate among the three treatments. Elliott et al. (1993) pointed out that the concentrations of acetate correspondingly decreased and propionate in reverse increased as the amount of fat supplemented in diet increased from 0 to 5%, by which the ratio of acetate:propionate was decreased consequently. In contrast, ratio of acetate:propionate in ruminal fluid was increased due to the increase of acetate and concomitant decrease of propionate as the level of dietary supplementation of protected fat increased in the range of 0 to 15% (Palmquist et al., 1986; Schauff and Clark, 1989). These observations suggest that protected fat may not affect the fermentation in rumen but overuse of tallow in rations will certainly be deleterious to ruminal fermentation. As the NDF was at 37.5% or more (Table 1) in this study, supplementation of 2.5% lard or Prilled fat in rations for Holstein cows would not affect the production of ruminal VFA and would be of efficacy in increase of milk fat percentage.

Butyrate, valerate and isovalerate were not significantly different among the treatments whereas isobutyrate molar percentage was the greatest (p<0.05) when cow fed with Prilled fat supplemented ration comparing to the other two rations. Cows fed with lard-supplemented ration had greater isobutyrate molar percentage than those in control (p<0.05).

The changes in molar percentage of isobutyrate along with the time points post feeding showed that the isobutyrate molar percentage, 1.59%, in PF was significantly lower than those in the other two treatments. The concentration of isobutyrate in ruminal fluid is a minor component of total VFA. Its level is usually less than 2% of total VFA. The changes in isobutyrate concentrations

Table 4. Effect of dietary supplementation of fat on pH value, and concentrations of NH3-N and VFAs in rumen fluid of Holstein dry cows (Ruminal trial)

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Control (3)</th>
<th>LD (3)</th>
<th>PF (3)</th>
<th>SEM</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>6.67</td>
<td>6.69</td>
<td>6.60</td>
<td>0.035</td>
</tr>
<tr>
<td>NH3-N, mg/dl</td>
<td>15.9</td>
<td>14.4</td>
<td>16.2</td>
<td>1.16</td>
</tr>
<tr>
<td>Total VFA, mM</td>
<td>105</td>
<td>111</td>
<td>108</td>
<td>5.3</td>
</tr>
<tr>
<td>VFA, mol/100 mol</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Acetate (A)</td>
<td>65.9</td>
<td>66.0</td>
<td>65.9</td>
<td>0.85</td>
</tr>
<tr>
<td>Propionate (P)</td>
<td>17.6</td>
<td>18.8</td>
<td>18.5</td>
<td>0.85</td>
</tr>
<tr>
<td>A:P</td>
<td>3.78</td>
<td>3.53</td>
<td>3.58</td>
<td>0.204</td>
</tr>
<tr>
<td>Butyrate</td>
<td>10.9</td>
<td>9.8</td>
<td>10.5</td>
<td>0.61</td>
</tr>
<tr>
<td>Isobutyrate</td>
<td>1.59c</td>
<td>1.65b</td>
<td>1.77a</td>
<td>0.009</td>
</tr>
<tr>
<td>Valerate</td>
<td>1.36</td>
<td>1.19</td>
<td>1.13</td>
<td>0.092</td>
</tr>
<tr>
<td>Isovalerate</td>
<td>2.59</td>
<td>2.59</td>
<td>2.26</td>
<td>0.196</td>
</tr>
</tbody>
</table>

Control was fed with basal ration, LD basal ration supplemented with 2.5% lard, and PF basal ration supplemented with 2.5% Prilled fat.

Experimental unit: each experimental period in each cow was regarded as an experimental unit.

a,b,c Means within the same row without the same superscripts are significantly different (p<0.05).
among the treatments were therefore not likely an important issue.

This study found that dietary supplementation of 2.5% lard or Prilled fat for dry Holstein cows did not raise adverse effects on their ruminal fermentation. The similar results have been reported by Grummer (1988), who used a basal diet comprised of corn, alfalfa hay and corn silage supplemented with fat as Ca soap or Prilled fat and fed the rations to lactating cows, found that the molar percentages of acetate and propionate or the ratio of acetate:propionate were not influenced by dietary supplementation of fat. It is concluded that either LD or PF has had no deleterious effects on rumen fermentation. However, the readers have to bear this in mind that these results observed in dry cows might not occur similarly in lactating cows.

CONCLUSION

The results of this research demonstrate that dietary supplementation of 2.5% prilled fat or lard increases plasma concentrations of non-esterified fatty acids without adversely affecting ruminal fermentation. This is especially true for the tropical and subtropical areas where heat stress is always concerned as an important issue. Supplementation of Prilled fat at 2.5% in rations for lactating cows in early lactation increased their milk yield and milk fat percentage accompanying with no apparent effects on the other milk compositions. Supplementation of lard at 2.5% in rations for lactating cows did increase milk fat percentage. These results suggest that supplementing fat to rations for cows in early lactation effectively increases milk fat percentage. It is especially attractive to dairy farmers when the price of raw milk is positive weighted by the increase of milk percentage, especially attractive to dairy farmers when the price of raw milk is positive weighted by the increase of milk percentage, like does in Taiwan currently.

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