

# Effect of different Roughage: Concentrate Ratio on Milk Yield and Its Fatty Acid Profile in Dairy Cows

Netsanet Beyero<sup>1\*</sup>, V. Kapoor<sup>2</sup>, B.S. Tewatia<sup>2</sup>

1, Department of Animal and Range Sciences, College of Agriculture, Wolaita Sodo University, Wolaita, Ethiopia.

2, College of Animal Science, Lala Lajpat Rai University of Veterinary and Animal Science, Hisar, India.

## Abstract

Experiment was conducted on eighteen crossbred dairy cows divided into three groups of six animals in each group. Cows were fed on roughage (berseem hay, green sorghum forage and wheat straw) and concentrate. The ratio of roughage: concentrate varied in the three treatment groups from 50:50 (T<sub>1</sub>); 60:40 (T<sub>2</sub>); and 70:30 (T<sub>3</sub>). DMI decreased with increasing the level of roughage in the diet. Digestibility of DM, CP, EE, NFE, and OM showed a significant decrease (P <0.05) when R:C was increased. NDF digestibility did not vary between treatment groups. On the other hand, CF and ADF digestibility were significantly (P <0.05) decreased with decreasing R:C. Increasing R:C significantly (P <0.05) decreased milk production, but production of 4% FCM was not affected by forage level. Milk fat concentration was significantly (P <0.05) decreased by 6.30 and 3.78 % in T<sub>1</sub> and T<sub>2</sub>, respectively, as compared T<sub>3</sub>. Increasing R:C had non significant effect on milk lactose, CP, TS, SNF, MUN, Ca and P level. Milk cholesterol level in T<sub>3</sub> was lowered by 13.35 and 8.27% as compared to T<sub>1</sub> and T<sub>2</sub>, respectively. The concentrations of TLCFA, TUFA, MUFA and PUFA were increased with the increase in roughage level in the diet, whereas, TSFA level was reduced. PUFA content of milk fat (g/100g fat) was increased by 141.75% in T<sub>3</sub> over T<sub>1</sub>. Total CLA production (mg/g fat) in T<sub>3</sub> was increased by 50.92% over T<sub>1</sub>. Total feeding cost decreased as the level of roughage in the ration increased. Total feeding cost decreased as the level of roughage in the ration increased. Feeding cost of T<sub>3</sub> per g PUFA production reduced by 68% and 48% as compared to T<sub>1</sub> and T<sub>2</sub>, respectively. Feeding cost of T<sub>3</sub> per g CLA production was reduced by 47% as compared to T<sub>1</sub>. The feed cost per kg milk, per g PUFA and CLA production was lowest for the cows fed 70:30 roughage: concentrate ratio diet. In summary, these results suggest that cows producing 12-14kg of milk/d could be best maintained on a dietary regimen of 70% good quality roughage and 30% concentrate to have the maximum production of human health beneficial conjugated linoleic acid without compromising on milk yield.

**Keywords:** Dairy cow, concentrate, roughage, milk fatty acid, conjugated linoleic acid

## 1. INTRODUCTION

Milk fat contains a substantial concentration of saturated fatty acids (C<sub>14:0</sub> and C<sub>16:0</sub>) and relatively low concentrations of monounsaturated and polyunsaturated fatty acids (Kennelly and Glimm, 1998). The ideal milk fat would contain 10 percent polyunsaturated fatty acids (PUFA), 8 percent saturated fatty acids (SFA), and 82 percent monounsaturated fatty acids (MUFA). The difference in fatty acid composition between the ideal and the actual milk fats is enormous. Increasing MUFA and PUFA at the expense of saturated fatty acids (C<sub>14:0</sub> and C<sub>16:0</sub>) is desirable from a human health perspective. Human dietary recommendations indicate the need to decrease intake of medium-chain saturated fatty acids (lauric acid, myristic acid and palmitic acid) to reduce the frequency of cardio-vascular diseases. Moreover, an increased dietary intake of polyunsaturated fatty acids stimulates the immune system and reduces the frequency of cancer and cardio-vascular diseases (Parodi, 1994; Huang *et al.*, 2008).

CLA is an intermediary product of ruminal biohydrogenation of dietary lipids. The biohydrogenation in the rumen that can alter the CLA production in milk fat is affected by the type and the amount of fatty acid substrate (Noble *et al.*, 1974; Halima *et al.*, 2008), and forage to grain ratio (Gerson *et al.*, 1985; Halima *et al.*, 2008) in the diet. The PUFA content of milk fat is an important aspect of establishing its dietetic quality. The nutritional value of milk fat can be improved by pasture feeding (Dhiman *et al.*, 1999), changing the forage-to-concentrate ratio to reduce proportions of saturated fatty acids and enhance the proportions of oleic, linoleic and linolenic fatty acids in bovine ration and increased ruminal production of CLA and its secretion into milk fat (Sutton and Morant, 1989). This study was designed to assess the effect of various roughage: concentrate ratios on milk yield and its fatty acid composition.

## 2. MATERIALS AND METHODS

### Experimental animals and management

The study was conducted on the Animal farm of College of Veterinary Sciences, Lala Lajpat Rai University of Veterinary and Animal Sciences (LLRUVAS), Hisar, India. Eighteen Cross bred dairy cattle in second to fourth lactation, with average body weight of 395±16.43kg and average daily milk yield 11.65±1.86kg were divided into four dietary groups of six in each group on the basis of milk yield and lactation stage. All the experimental

animals were housed in well ventilated shed and maintained under clean and hygienic condition in the barn of having the arrangement of individual feeding. Fresh and clean tap water was provided free choice to all animals. The experimental animals were given twenty-one days adaptation period. Before starting the experiment, the animals were adapted for 21 days to the diets. The duration of experiment was 60 days from July to September 2011.

### Experimental diets and treatments

The experimental animals were kept on diets containing concentrate mixture (Table 2.1) and roughage (berseem hay, sorghum green forage and wheat straw). Berseem hay was chopped manually to approximately 5-7 cm length before feeding. Sorghum green forage was harvested daily, and chopped manually also to approximately 5-7 cm length and mixed before feeding. Animals were fed to meet the nutrient requirement (NRC, 2001). All the treatments were iso-nitrogenous and iso-caloric. The three treatment groups differed with regard to the different proportion of roughage to concentrate in the diet. The ratio of roughage: concentrate varied from 50:50 (T<sub>1</sub>); 60:40 (T<sub>2</sub>) and 70:30 (T<sub>3</sub>).

**Table 2.1 Percent composition of ingredients in concentrate mixture**

Ingredients	Parts/100kg
Groundnut cake	30
Wheat	27
Barely	40
Mineral mixture	2
Common salt	1

### Data collection

The experimental feedstuffs were weighed before feeding. Feed refusals were collected from individual animals every morning and weighed. Feed offered and refusals were sampled daily, bulked for each animal and sub-sampled for chemical analysis. Amount of all feedstuffs (Berseem forage, wheat straw and concentrate mix) offered and refused were measured to quantify feed intake. Animals were hand milking twice a day at 4 a.m. and 5 p.m. during the experimental period and milk yield was recorded by using circular dial type spring balance, with the capacity of 20 kg and an accuracy of  $\pm 0.05$  kg, at each milking time. Milk samples for analysis were taken from morning and evening milking at weekly interval. Hundred fifty ml of milk sample was taken from each milking and each animal in milk sampling bottles. The milk samples from morning and evening samples were pooled and milk was thoroughly mixed before analysis.

### Digestibility trial

A digestibility trial of five days collection period was conducted at the end of experimental period to determine the digestibility of nutrients. The feeding regime during the digestibility trial was the same as in the feeding experiment. During trials, the sample of feed offered, residue left and faeces voided were collected and the representative samples of daily feeds offered, residue left and faeces voided were collected and oven dried. For nitrogen estimation, 1/600<sup>th</sup> part of the total faeces was weighed and preserved in 40% H<sub>2</sub>SO<sub>4</sub>, in wide mouthed plastic bottles. At the end of the trial, samples from each animal were pooled, mixed and grounded to 1mm sieve size. The ground samples were stored for further analysis.

### Chemical analysis

Samples of feeds offered, refusals and faeces were analyzed for dry matter (DM), total ash, crude protein (CP), ether extract (EE) and crude fibre (CF) (AOAC, 2005), neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) contents (Goering and Van Soest, 1970). The calcium content of feed samples was determined by the atomic absorption spectrophotometry and Phosphorus concentration was determined by colorimetric method.

Samples of milk were analyzed for total solid, total fat and protein according to AOAC, 2005. Milk lactose content (Perry and Doon, 1950), cholesterol (Bindal and Jain, 1973), milk urea nitrogen (MUN) (Bector *et al.*, 1998), Fatty acid composition (Sukhija and Palmquist, 1984 modified by Dhiman *et al.*, 1999), CLA content (AOAC, 1995).

### Statistical analysis

The data were subjected to analysis of variance using SPSS statistical software version 16.0 for Windows. The means which showed significant differences at the probability level of  $P < 0.05$  were compared with each other by using LSD.

### 3. RESULTS AND DISCUSSION

#### 3.1 Chemical composition of experimental diets

The chemical composition of the diets is presented in Table 3.1.

**Table 3.1 Chemical composition of dietary ingredients (concentrate mixture, sorghum forage, berseem hay and wheat straw)**

Item	Concentrate mixture	Sorghum forage	Berseem hay	Wheat straw
DM	91.27	31.09	87.81	90.35
OM	91.40	91.18	88.87	93.27
NFE	57.60	53.44	43.78	47.15
CP	19.10	7.82	12.60	2.10
EE	6.55	1.70	1.66	1.55
CF	8.15	28.22	30.83	41.97
Ash	8.60	8.82	11.13	6.73
NDF	28.22	63.70	56.67	79.00
ADF	10.47	41.10	38.43	49.85
ADL	4.69	7.60	12.63	12.55
Calcium	1.36	0.64	1.47	0.31
Phosphorus	0.77	0.26	0.32	0.10

#### 3.2 Dry matter intake, Nutrient intake and Nutrient digestibility

Dry matter intake significant ( $P < 0.05$ ) increased with the decrease in the level of roughage in the diet (Table 3.2). The largest increase was between 50:50 and 60:40 roughage to concentrate (R:C) ratio diets. This is similar to the response of DMI observed by Soita *et al.* (2005) who reported that feeding a higher level concentrate (R:C ratios 45:55) diet ( $P < 0.05$ ) increased DMI by 1.6 kg/d than diet containing lower proportion of concentrate (55:45 roughage to concentrate ratio). Macleod *et al.* (1983) also reported a constant increase in DMI until forage was reduced to 35% of the diet. The lower DMI with high roughage diet may be due to limited gut fill capacity in the diets rich in roughage. The intakes of DCP and TDN varied significantly ( $P < 0.05$ ) among treatment groups and the values were highest in T<sub>1</sub> ( $0.95 \pm 0.01$  and  $7.25 \pm 0.03$  kg/d) and lowest in T<sub>3</sub> ( $0.88 \pm 0.02$  and  $6.85 \pm 0.14$  kg/d). Increasing roughage: concentrate ratio in the diet of cows significantly ( $P < 0.05$ ) reduced DCP and TDN intakes. Further, studies of Gaafar *et al.* (2009) and Macleod *et al.* (1983) also reported that feeding high concentrate (R:C ratio 40:60) diets to buffalos substantially increased DCP and TDN intakes by 30 and 12%, respectively over high roughage (R:C ratio 60:40) diet. In the current study, the increase in DCP and TDN intakes in high concentrate (R:C ratio 50:50) diet was 7.5 and 5.5% over low concentrate (R:C ratio 70:30) diet. This may be attributed to the higher DMI in T<sub>1</sub> as compared to T<sub>2</sub> and T<sub>3</sub> diets.

**Table 3.2 Dry matter intake (DMI), nutrient intake and digestibility of nutrients of cows fed diets having different ratios of roughage: concentrate**

Attribute	T <sub>1</sub> (R:C 50:50)	T <sub>2</sub> (R:C 60:40)	T <sub>3</sub> (R:C 70:30)
Feed Intake (kg/d)			
Roughage	$6.89 \pm 0.25^c$	$7.50 \pm 0.31^b$	$8.61 \pm 0.24^a$
Concentrate mixture	$7.30 \pm 0.00^a$	$5.93 \pm 0.00^b$	$4.56 \pm 0.00^c$
Total DMI (kg)	$14.19 \pm 0.25^a$	$13.43 \pm 0.31^{ab}$	$13.18 \pm 0.24^b$
DMI (% Body Wt)	$3.70 \pm 0.06^a$	$3.48 \pm 0.10^{ab}$	$3.36 \pm 0.02^b$
DMI (g/ W <sup>0.75</sup> kg)	$168.08 \pm 2.84^a$	$153.69 \pm 3.49^b$	$149.22 \pm 1.71^b$
Nutrient Intake (kg/d)			
DCP	$0.95 \pm 0.01^a$	$0.92 \pm 0.01^{ab}$	$0.88 \pm 0.02^b$
TDN	$7.25 \pm 0.03^a$	$7.07 \pm 0.10^{ab}$	$6.85 \pm 0.14^b$
Nutrient Digestibility (%)			
DM	$63.71 \pm 0.37^a$	$61.60 \pm 0.29^{ab}$	$60.03 \pm 1.20^b$
OM	$65.52 \pm 0.32^a$	$63.97 \pm 1.02^{ab}$	$62.07 \pm 1.50^b$
NFE	$68.61 \pm 0.77^a$	$65.72 \pm 0.98^{ab}$	$63.56 \pm 1.62^b$
CP	$71.13 \pm 0.44^a$	$69.58 \pm 0.58^{ab}$	$67.72 \pm 1.18^b$
EE	$65.42 \pm 0.67^a$	$62.82 \pm 0.7^a$	$58.12 \pm 1.14^b$
CF	$51.73 \pm 0.67^b$	$55.06 \pm 0.60^a$	$54.46 \pm 0.32^a$
NDF	$53.54 \pm 1.94$	$54.14 \pm 1.42$	$55.72 \pm 4.5$
ADF	$46.00 \pm 0.86^b$	$47.03 \pm 1.15^b$	$51.31 \pm 0.85^a$

Values bearing different superscripts in a row differ significantly ( $P < 0.05$ ).

Increasing the proportion of forage in the diet from 50 to 70% decreased ( $P < 0.05$ ) apparent digestibilities of DM and OM. Higher digestibility of DM and OM in the total tract with low roughage to concentrate ratio agreed with earlier reports (Rode *et al.*, 1985; Sarwar *et al.*, 1992 and Yang *et al.*, 2001) indicating decreased apparent digestibility of DM and OM in the total tract as a result of increase in the proportion of roughage in diet from 35 to 65%. Digestibility of NDF fractions of the diets in treatments tended to be lower when the roughage proportion was increased in the diet, though the differences were non significant. The result of (Kalscheur *et al.*, 1997) indicated that digestibility of NDF was lower ( $P < 0.007$ ) for cows fed low fibre (R:C ratio 25:75) diets than for cows fed high fibre (F:C ratio 60:40) diets. In contrast, Yang *et al.* (2001) observed lower digestibility of NDF by increasing the R:C ratio from 35:65 to 65:35. Apparent digestibility of ADF and CF was significantly lower ( $P < 0.05$ ) in  $T_1$  than in  $T_3$ . Similar depression of fiber digestibility with high concentrate diets was reported when the percentage of forage in the diet decreased from 60 to 25% (Rode *et al.*, 1985, Kalscheur *et al.*, 1997 and Moorby *et al.*, 2006). The decrease in ruminal fiber digestion in high concentrate diet is believed to be caused by the inhibited growth of cellulolytic bacteria when ruminal pH decreases below 6.2 (Grant and Mertens, 1992).

### 3.3 Milk yield and its composition

**Table 3.3 Milk yield and milk composition of cows fed diets having different ratios of roughage: concentrate**

Attribute	$T_1$ (R:C 50:50)	$T_2$ (R:C 60:40)	$T_3$ (R:C 70:30)
Milk yield (kg/d)	13.78±0.45 <sup>a</sup>	12.96±0.43 <sup>ab</sup>	12.28±0.52 <sup>b</sup>
FCM (kg/d)	13.21±0.42	12.67±0.47	12.23±0.53
Fat yield (kg/d)	0.51±0.02	0.50±0.02	0.49±0.02
Milk composition (%)			
Total solids	11.11±0.52	11.31±0.27	11.14±0.34
Crude protein	3.14±0.04	3.04±0.08	3.01±0.11
Fat	3.72±0.03 <sup>c</sup>	3.85±0.04 <sup>b</sup>	3.97±0.04 <sup>a</sup>
Lactose	4.50±0.02	4.46±0.02	4.45±0.03
SNF	7.38±0.53	7.46±0.26	7.16±0.33
Total ash	0.74±0.04	0.72±0.01	0.74±0.01
Calcium	0.108±0.003	0.106±0.004	0.106±0.002
Phosphorus	0.062±0.001	0.063±0.001	0.062±0.002
MUN (mg/dl)	12.74±0.12	12.58±0.08	12.11±0.05
Cholesterol (mg/100ml)	6.03±0.06 <sup>a</sup>	5.76±0.11 <sup>a</sup>	5.32±0.10 <sup>b</sup>

Values bearing different superscripts in a row differ significantly ( $P < 0.05$ ).

Increasing roughage: concentrate ratio significantly ( $P < 0.05$ ) decreased milk production (Table 3.3), but production of 4% FCM was not affected by forage level, because milk fat content was inversely related to milk production. Increased milk production with increased concentrate feeding has been consistently reported in lactating dairy cows. Macleod *et al.* (1983) who observed an increase of 0.06 kg milk per percentage unit increase of concentrates, while the increase in current study averaged 0.075 kg milk per percentage unit increase of concentrates. A similar increase in milk yield with increased dietary concentrate has been reported by other workers, (Tuan, 2000; Sanh *et al.*, 2002 and Kuoppala *et al.*, 2004). This may be attributed to increased DM and energy intake. The lack of effect of R:C ratio on FCM production is also in agreement with earlier reports (Beauchemin and Beauchemin-Smith, 1989; Beauchemin *et al.*, 1994). Increasing roughage: concentrate in the diet of cows increased milk fat production, confirming results of other studies (Loor *et al.*, 2005) in which it was observed that feeding high concentrate (R:C ratio 35:65) diets substantially reduced milk fat percentage (28%) as compared to R:C ratio 65:35 diet. In the current study the reduction was 6.30 and 3.78 % in  $T_1$  and  $T_2$ , respectively, as compared to  $T_3$ . Similarly, Kalscheur *et al.* (1997) observed reduction ( $P < 0.01$ ) in milk fat (3.67%) of cows fed low fibre (F:C ratio 25:75) diets compared with cows fed high fibre (F:C ratio 60:40) diets (4.16%). This effect has been commonly reported for high concentrate diets (Macleod *et al.*, 1983; Hansen *et al.*, 1991; Yang *et al.*, 2001 and Moorby *et al.*, 2006) and is clearly an indication of milk fat depression due to ruminal processes (Van Soest, 1963; Bauman *et al.*, 2008). Forage diets high in cellulose give rise to acetic acid, while concentrate diets give rise to propionic acid thereby reducing the proportion of acetic acid (McDonald *et al.*, 2002). High levels of concentrate are conducive to production of propionic acid in the rumen, which in turn promotes partition of energy towards synthesis of body fat instead of milk fat synthesis, resulting in a decrease in milk fat content (Randby, 1996; Sanh *et al.*, 2002). In the present study, protein and lactose percentages in milk did not vary ( $P < 0.05$ ) as roughage: concentrate was increased from 50:50 to 70:30 ratios. A number of earlier reports support this findings (Broster *et al.*, 1985; Sutton *et al.*, 1987 and Sutton, 1989).

It is evident from the present study that dietary R:C ratios did not influence the milk MUN, calcium and phosphorus contents. Cows fed T<sub>3</sub> diets had significantly ( $P < 0.05$ ) lower ( $5.32 \pm 0.10$ ) milk cholesterol level than cows in T<sub>1</sub> ( $6.03 \pm 0.06$ ), and T<sub>2</sub> ( $5.76 \pm 0.11$ ) treatment groups. The cholesterol production value in T<sub>3</sub> was lowered by 13.35 and 8.27 % as compared to T<sub>1</sub> and T<sub>2</sub>, respectively. Medium chain fatty acids namely, lauric acid (C<sub>12:0</sub>), myristic acid (C<sub>14:0</sub>) and palmitic acid (C<sub>16:0</sub>) are considered to be cholesterol-raising fatty acids (Berner, 1993; Aloka, 1997). In the current study, the content of each of these fatty acids in milk was significantly ( $P < 0.05$ ) increased when the level of concentrate increased in the diets, and the increased level of cholesterol with increasing concentrate in the diet had similar trends with those fatty acids.

### 3.4 Fatty acid intake and milk fatty acid profile

#### i. Fatty acid composition of experimental diets

**Table 3.4 Fatty acid composition (mg/g DM) of dietary ingredients**

Fatty acids	Concentrate mixture	Sorghum forage	Berseem hay	Wheat straw
C <sub>8:0</sub>	0.081	0.045	0.124	0.031
C <sub>10:0</sub>	0.047	0.034	0.158	0.013
C <sub>12:0</sub>	0.068	0.068	0.245	0.036
C <sub>14:0</sub>	0.127	0.078	0.112	0.058
C <sub>14:1</sub>	0.314	ND	0.168	ND
C <sub>16:0</sub>	12.21	0.312	5.156	0.506
C <sub>16:1</sub>	0.315	0.049	0.632	0.039
C <sub>18:0</sub>	15.544	0.122	2.344	0.254
C <sub>18:1</sub>	17.662	0.468	2.768	0.404
C <sub>18:2</sub>	14.048	0.446	3.065	0.388
C <sub>18:3</sub>	2.558	0.095	7.26	0.078
C <sub>20:0</sub>	0.665	0.018	0.542	0.022
Total fatty acids	63.639	1.735	22.574	1.829
Total saturated fatty acids	28.742	0.677	8.681	0.92
Total unsaturated fatty acids	34.897	1.058	13.893	0.909
Total long chain fatty acids	63.002	1.51	21.767	1.691
Monounsaturated fatty acids	18.291	0.517	3.568	0.443
Polyunsaturated fatty acids	16.606	0.541	10.325	0.466

ND- Not detectable

The composition of fatty acids (g/g DM) in the dietary ingredients has been presented in Table 3.4. Result on Table 3.4 indicated that linoleic (C<sub>18:2</sub>) and linolenic (C<sub>18:3</sub>) acids when consider together contributed 45.74% and 31.18% of the total fatty acids in berseem hay and sorghum forage, respectively while total contribution of these fatty acids towards concentrate mixture fat was 26.09%. Total saturated fatty acids concentration (mg/g DM) in concentrate mixture was 28.742 and the corresponding values for berseem hay and sorghum forage were 0.677 and 8.681 respectively. The data revealed that concentration of Total unsaturated fatty acids (TUFA) accounted 54.69, 60.98 and 61.54% of total fat in concentrate, berseem hay and sorghum forage, respectively.

#### ii. Fatty acids intake

Feeding high concentrate diet (T<sub>1</sub>) increased ( $P < 0.05$ ) total fatty acid intake by 49.7 and 83.71g/d as compared to T<sub>2</sub> and T<sub>3</sub>, respectively (Table 3.5). The intake of oleic (C<sub>18:1</sub>) and linoleic (C<sub>18:2</sub>) acid was also were greater ( $P < 0.05$ ) in response to high concentrate (T<sub>1</sub>) diet; however linolenic (C<sub>18:3</sub>) acid intake was lower (14.1 and 23.33g/d) with T<sub>1</sub> vs. T<sub>2</sub> and T<sub>3</sub>, respectively. A significantly higher intake of oleic and linoleic acid in T<sub>1</sub> was reflected on TUFA intake. Similarly, Loo *et al.* (2004) reported that feeding high concentrate diet increased total fatty acid intake by 109g/d, and oleic and linoleic acid intakes were also greater in response to higher concentrate (35:65 ratio of R:C) diets, however linolenic acid intake was lower by 26.6 g/d with high concentrate diet as compared to low concentrate high roughage (65:35 ratio of R:C) diets. High intake of oleic and linoleic acid in T<sub>1</sub> cows is a reflection of higher level of those fatty acids in concentrate (Table 3.4). MUFA intake followed a similar trend like oleic acid intake, as oleic acid contributed about 95% of the total MUFA. Daily intake of PUFA intake ranged between  $125.13 \pm 0.85$  (T<sub>3</sub>) to  $128.48 \pm 1.00$  (T<sub>1</sub>).



**Table 3.5 Fatty acids intake of cows fed diets different ratios of roughage: concentrate**

Fatty acid Intake (g/d)	T <sub>1</sub> (R:C 50:50)	T <sub>2</sub> (R:C 60:40)	T <sub>3</sub> (R:C 70:30)
C <sub>8:0</sub>	0.96±0.02 <sup>b</sup>	1.01±0.02 <sup>b</sup>	1.15±0.01 <sup>a</sup>
C <sub>10:0</sub>	0.64±0.02 <sup>c</sup>	0.81±0.02 <sup>b</sup>	1.09±0.01 <sup>a</sup>
C <sub>12:0</sub>	1.07±0.03 <sup>c</sup>	1.32±0.03 <sup>b</sup>	1.75±0.02 <sup>a</sup>
C <sub>14:0</sub>	1.54±0.02 <sup>a</sup>	1.45±0.02 <sup>b</sup>	1.46±0.01 <sup>b</sup>
C <sub>14:1</sub>	2.35±0.01 <sup>a</sup>	2.21±0.02 <sup>b</sup>	2.19±0.01 <sup>b</sup>
C <sub>16:0</sub>	93.15±0.53 <sup>a</sup>	85.06±0.56 <sup>b</sup>	80.57±0.43 <sup>c</sup>
C <sub>16:1</sub>	2.86±0.06 <sup>c</sup>	3.48±0.07 <sup>b</sup>	4.54±0.05 <sup>a</sup>
C <sub>18:0</sub>	115.18±0.24 <sup>a</sup>	97.83±0.25 <sup>b</sup>	82.14±0.19 <sup>c</sup>
C <sub>18:1</sub>	133.31±0.32 <sup>a</sup>	113.35±0.33 <sup>b</sup>	95.40±0.25 <sup>c</sup>
C <sub>18:2</sub>	106.85±0.35 <sup>a</sup>	92.40±0.36 <sup>b</sup>	80.14±0.27 <sup>c</sup>
C <sub>18:3</sub>	21.63±0.66 <sup>c</sup>	30.89±0.76 <sup>b</sup>	44.99±0.58 <sup>a</sup>
C <sub>20:0</sub>	5.16±0.05 <sup>b</sup>	5.18±0.06 <sup>b</sup>	5.58±0.04 <sup>a</sup>
Total saturated fatty acids	217.71±0.90 <sup>a</sup>	192.67±0.96 <sup>b</sup>	173.73±0.73 <sup>c</sup>
Total unsaturated fatty acids	267.00±1.40 <sup>a</sup>	242.34±1.54 <sup>b</sup>	227.26±1.17 <sup>c</sup>
Total long chain fatty acids	478.14±2.21 <sup>a</sup>	428.20±2.40 <sup>b</sup>	393.34±1.82 <sup>c</sup>
Monounsaturated fatty acids	138.52±0.40 <sup>a</sup>	119.05±0.42 <sup>b</sup>	102.13±0.32 <sup>c</sup>
Polyunsaturated fatty acids	128.48±1.00 <sup>a</sup>	123.29±1.12 <sup>b</sup>	125.13±0.85 <sup>b</sup>

Values bearing different superscripts in a row differ significantly ( $P < 0.05$ ).

### iii. Fatty acid profile of milk fat

Fatty acid profile of milk of cows under experimental feeding is shown in Table 3.6. The Table revealed that as the dietary roughage proportion increased there was a significant ( $P < 0.05$ ) reduction in the concentration (g/100g fat) of C<sub>10:0</sub>, C<sub>12:0</sub>, C<sub>14:0</sub> and C<sub>16:0</sub> fatty acids. Lower percentages of these fatty acids in cows fed high fiber diet were also reported by AlZahal *et al.* (2008). Similarly, Martini *et al.* (2010) also observed decreased values of some medium chain fatty acids viz. C<sub>12:0</sub> (14.89%) and C<sub>14:0</sub> (4.03%) with the increase in roughage level in the diet of lactating ewes. On the other hand, an inverse effect ( $P < 0.05$ ) was observed in relation to the palmitoleic (C<sub>16:1</sub>), stearic (C<sub>18:0</sub>), oleic (C<sub>18:1</sub>), linoleic (C<sub>18:2</sub>), and linolenic (C<sub>18:3</sub>) acids, which increased with the level of increased roughage in the diet. Even if linoleic acid intake was significantly ( $P < 0.05$ ) higher in T<sub>1</sub>, milk linoleic acid was significantly lower, while milk linolenic acid was increased with intake (Table 3.5 and Table 3.6). A high level of milk saturated fatty acid and lower linoleic acid concentration in T<sub>1</sub> may be explained on the basis that ruminal biohydrogenation of unsaturated FA was comparatively more complete in cows fed linoleic acid than in cows fed linolenic acid.

**Table 3.6 Fatty acid composition (g/100g) of milk fat of cows fed diets having different ratios of roughage: concentrate**

Fatty acids	T <sub>1</sub> (R:C 50:50)	T <sub>2</sub> (R:C 60:40)	T <sub>3</sub> (R:C 70:30)
C <sub>8:0</sub>	0.96±0.05	0.94±0.05	1.13±0.09
C <sub>10:0</sub>	2.48±0.15 <sup>a</sup>	2.18±0.10 <sup>ab</sup>	2.02±0.15 <sup>b</sup>
C <sub>12:0</sub>	3.21±0.12	3.03±0.13	3.03±0.23
C <sub>14:0</sub>	13.26±0.24 <sup>a</sup>	10.77±0.17 <sup>b</sup>	9.23±0.15 <sup>c</sup>
C <sub>14:1</sub>	1.44±0.09 <sup>b</sup>	1.70±0.13 <sup>ab</sup>	1.78±0.08 <sup>a</sup>
C <sub>16:0</sub>	26.80±2.06	26.37±0.32	25.24±0.23
C <sub>16:1</sub>	2.04±0.12 <sup>b</sup>	2.53±0.13 <sup>a</sup>	2.46±0.12 <sup>a</sup>
C <sub>18:0</sub>	17.59±0.23 <sup>a</sup>	16.91±0.12 <sup>b</sup>	16.16±0.20 <sup>b</sup>
C <sub>18:1</sub>	23.58±0.33 <sup>c</sup>	27.01±0.15 <sup>b</sup>	28.30±0.20 <sup>a</sup>
C <sub>18:2</sub>	2.08±0.07 <sup>c</sup>	3.28±0.08 <sup>b</sup>	4.13±0.08 <sup>a</sup>
C <sub>18:3</sub>	1.01±0.05 <sup>c</sup>	1.94±0.11 <sup>b</sup>	3.34±0.27 <sup>a</sup>
C <sub>20:0</sub>	0.89±0.06 <sup>a</sup>	0.65±0.06 <sup>b</sup>	0.71±0.06 <sup>b</sup>
Total saturated fatty acids	65.19±2.15 <sup>a</sup>	60.86±0.59 <sup>b</sup>	57.53±0.28 <sup>c</sup>
Total unsaturated fatty acids	30.16±0.29 <sup>c</sup>	36.46±0.36 <sup>b</sup>	40.01±0.21 <sup>a</sup>
Total long chain fatty acids	74.00±2.09 <sup>b</sup>	78.70±0.42 <sup>a</sup>	80.35±0.33 <sup>a</sup>
Monounsaturated fatty acids	27.06±0.35 <sup>c</sup>	31.24±0.27 <sup>b</sup>	32.55±0.20 <sup>a</sup>
Polyunsaturated fatty acids	3.09±0.08 <sup>c</sup>	5.22±0.14 <sup>b</sup>	7.47±0.21 <sup>a</sup>
Total CLA (mg/g fat)	10.31±0.28 <sup>c</sup>	12.10±0.35 <sup>b</sup>	15.56±0.33 <sup>a</sup>

Values bearing different superscripts in a row differ significantly ( $P < 0.05$ ).

Total long chain fatty acids (TLCFA), total unsaturated fatty acids (TUFA), monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA) increased significantly ( $P < 0.05$ ) with the increased proportion of roughage in the diet (Table 3.6). Total MUFA content varied from 28.38 ( $T_1$ ) to 33.37 ( $T_3$ ) % of milk fat. The concentration of PUFA was highest in  $T_3$  (7.66 % of milk fat) and lower in  $T_1$  (3.24 % of milk fat). Polyunsaturated fatty acids content of milk fat (g/100g fat) was increased by 68.93 and 141.75% in  $T_2$  and  $T_3$ , respectively over  $T_1$ . Polyunsaturated fatty acids are not synthesized by the tissue of ruminants, so their concentration in the milk strictly depends on the amount of the fat absorbed in the intestines and, therefore, on the amounts released in the rumen. Therefore, the increase in the roughage level proportionally decreases the concentrate contents and hence the availability of unsaturated fatty acids to be used by the mammary gland in the synthesis of milk lipids (Grummer, 1991; Chilliard *et al.*, 2001b and Mesquita *et al.*, 2008). Dairy cows on herbage-based diets derive fatty acids for milk fat synthesis from the diet and rumen microorganisms (400–450 g/kg), from adipose tissues ( $< 100$  g/kg), and from *de novo* biosynthesis in the mammary gland (about 500 g/kg) (Kalač and Samkova, 2010). The relative contributions of these FA sources to milk fat production are highly dependent upon feed intake, and diet composition. High intake of concentrate diet is associated with a higher level of *de novo* synthesis resulting in more saturated milk fat. In contrast, higher intakes of PUFAs from forage result in higher concentrations of oleic acid, vaccenic acid and CLA in milk fat (Walker *et al.*, 2004).

Concentrations of CLA were significantly ( $P < 0.05$ ) affected by dietary treatment varying in the proportion of roughage: concentrate (Table 3.6). Total CLA yield (mg/g fat) averaged  $10.31 \pm 0.28$ ,  $12.10 \pm 0.35$ , and  $15.56 \pm 0.33$  in  $T_1$ ,  $T_2$  and  $T_3$ , respectively. Total CLA content (mg/g fat) in  $T_3$  increased by 50.92 and 17.36% over  $T_1$  and  $T_2$ , respectively. Results from current study are in agreement with the previous reports of Tyagi *et al.* (2008) who reported that total CLA content averaged 18.0 and 6.6 mg/g in berseem and concentrate fed animals, respectively which showed 3 fold increases in total CLA content by green fodder feeding. Dhiman *et al.* (1999) also reported highest CLA content in the milk of cows fed solely on pasture than those fed on one third and two third pastures. Schroeder *et al.* (2003) recorded 2-3 fold higher milk CLA concentration in dairy cows fed pasture as compared with TMR.

#### 4.4 Economics of feeding

**Table 3.7 Economics of feeding for milk production and milk quality parameters in cows fed different ratios of roughage to concentrate**

Attribute	$T_1$	$T_2$	$T_3$
Cost of feeding concentrate mixture (USD/d)	2.04±0.00	1.66±0.00	1.27±0.016
Cost of feeding roughage (USD/d)	0.50±0.018 <sup>b</sup>	0.52±0.022 <sup>b</sup>	0.58±0.00 <sup>a</sup>
Total feeding cost (USD/d)	2.54±0.018 <sup>a</sup>	2.18±0.022 <sup>b</sup>	1.86±0.016 <sup>c</sup>
Total milk production (kg/d)	13.78 <sup>a</sup>	12.96 <sup>ab</sup>	12.28 <sup>b</sup>
Feeding cost/ kg milk production (USD)	0.18±0.005 <sup>a</sup>	0.17±0.006 <sup>ab</sup>	0.15±0.007 <sup>b</sup>
Total fat yield (kg/d)	0.513	0.500	0.488
Feeding cost/ kg fat yield (USD)	4.95±0.127 <sup>a</sup>	4.35±0.182 <sup>b</sup>	3.81±0.177 <sup>c</sup>
PUFA yield (g/100g fat)	3.09 <sup>c</sup>	5.22 <sup>b</sup>	7.47 <sup>a</sup>
PUFA yield (g/d)	15.84 <sup>c</sup>	26.08 <sup>b</sup>	36.31 <sup>a</sup>
Feeding cost/g PUFA (USD)	0.16±0.004 <sup>a</sup>	0.08±0.004 <sup>b</sup>	0.05±0.002 <sup>c</sup>
Total CLA yield (mg/g fat)	10.31 <sup>c</sup>	12.10 <sup>b</sup>	15.56 <sup>a</sup>
Total CLA yield (g/d)	5.27 <sup>c</sup>	6.03 <sup>b</sup>	7.56 <sup>a</sup>
Feeding cost/ g CLA (USD)	0.49±0.006 <sup>a</sup>	0.36±0.014 <sup>b</sup>	0.26±0.008 <sup>c</sup>

Values bearing different superscripts in a row differ significantly ( $P < 0.05$ ).

The feeding cost was lowest in group  $T_3$  cows fed on diet having 70% roughage and it was highest in group  $T_1$  in cows fed on the diet containing 50% roughage. The feeding cost (USD/d) was calculated by taking into account the cost of feed ingredients i.e. roughage and concentrate mixture. Total feeding cost was lowest (USD 1.86 /d) for cows fed on 70 % roughage, followed by the cows fed 60% roughage (USD 2.18/d) and (USD 2.54 /d) cows fed 50% roughage. Feeding cost per g PUFA production in group  $T_3$  was reduced by 68% and 48% as compared to  $T_1$  and  $T_2$ , respectively. Feeding cost per g CLA production was reduced by 47% in  $T_3$  as compared to  $T_1$ . The feed cost per kg milk produced, per g PUFA and per g CLA production was lowest for the cows fed roughage and concentrate in the ratio of 70:30. It was inferred that cows producing 12-14kg of milk/d could be best maintained on a dietary regimen of 70% roughage and 30% concentrate to have the maximum human health beneficial conjugated linoleic acid without compromising on milk yield.

#### CONCLUSION

In this experiment, we have demonstrated that berseem-based roughage feeding enhances the dietetic quality of

cow milk compared to concentrate-feeding. It may be inferred that cows producing 12-14kg of milk/d could be best maintained on a dietary regimen of 70% good quality roughage i.e. berseem hay and 30% concentrate to have the maximum production of human health beneficial fatty acids (PUFA including CLA) without compromising on milk yield.

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