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The Effects of Dietary ω -3 and ω -6 Fatty Acids on Nutrient Utilization and Growth Performance in Sahiwal Heifers

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ABSTRACT

This study was conducted to evaluate the effect of dietary supplementation with ω -3 and ω-6 fatty acid rich oil on nutrient utilization, growth performance and pubertal age in Sahiwal heifers. Eighteen healthy Sahiwal heifers of average 18.33±1.14 month's age and 194±4.16 kg body weights were randomly assigned to 3 treatment groups *viz* T_1, T_2 and T_3 with 6 animals in each group. Heifers were individually fed diets of chopped wheat straw, green fodder and concentrate mixture to meet nutrient requirement as per ICAR (2013). The concentrate mixtures fed to the heifers under treatment group T_2 and T_3 were supplemented with ω-6 fatty acid rich soybean oil and ω-3 fatty acid rich linseed oils @ 3.5 percent, respectively. To make the rations iso-caloric, palm oil @ 3.5 percent was supplemented in the concentrate mixture of heifers in control group (T_1). All the concentrate mixtures were isonitrogenous. Average daily DMI was significantly ($P<0.05$) higher in heifers fed linseed oil supplemented concentrate mixture as compared to others. Total body weight gain and ADG was significantly ($P<0.05$) higher T_3 than T_2 and control heifers. The OMD was significantly ($P<0.05$) improved in groups fed ω-6 and ω-3 fatty acid rich oils as compared to control. The CP, DCP and TDN percent; and TDN intake did not differ among different dietary treatments. Feeding of linseed oil resulted in significant ($P<0.05$) reduction in the estrous age of Sahiwal heifers. The total dry matter intake up to first estrous was reduced by 32.12 and 12.77 percent in T_3 and T_2, respectively, as compared to control (T_1). Similarly, total concentrate mixture intake up to attainment of heifers' pubertal age in ω-3 rich linseed oil and ω-6 rich soybean oil fed group reduced by 38.82 and 16.11 percent, respectively, over control diet. The study deduced that ω-3 fatty acid supplementation reduced pubertal age of Sahiwal heifers.

Keywords: ω-3 and ω-6 fatty acids, Growth, Nutrient utilization, Pubertal age, Sahiwal heifers

INTRODUCTION

Livestock rearing is an important source of employment, income and livelihood of rural population of India. Demand of milk is ever increasing over time. For maximum output from commercial dairy farming sound rearing approach of heifers so as to replace the older and unproductive animals through culling play a significant role. However, heifer production is considered to be the most expensive part of dairy farm operation because it needs more inputs for a longer period with no immediate returns. Proper planning for replacement heifers affects success and longevity of a dairy herd. For successful dairy farming, a complete package of heifer rearing is imperative to exploit the performance potential at minimum expense. As reported by Razzaque *et al.* (2010) the input in dairy industry in term of feed cost represents around 84% of the total input costs for heifer production. Heifers must reach 30 to 40% of their mature body weight in order to reach puberty. Adequate nutrient supply to the animal is paramount to development of heifers. Age at puberty is an important production trait; and hormones, nutrition, and genotype play key roles in attainment of puberty (Bellows and Hall, 1996).

High levels of grains in the concentrate mixture can adversely affect DMI, rumen fermentation and fiber digestibility. High level of dietary energy intake influences reproductive performance of animals, as reported by Barth *et al.* (2008) in young bulls through direct effect on the age at puberty and growth of scrotal circumference. Fat supplements can be used @ 20 to 30 g/kg DMI (Palmquist, 1988) without

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negatively affecting fibre digestibility.

The ω-6 and ω-3 FAs as well as their different dietary ratios can affect fermentation and nutrient digestibility in rumen (Zhang *et al.* 2008). Traditionally, lipids were added to the diet of bovine to increase the energy density and alleviate the negative energy balance during early lactation. But, recent studies have unveiled many beneficial effects of lipids other than providing energy, especially the one rich in polyunsaturated fatty acids. Some groups of FA are considered essential to mammals because mammalian cells are unable to synthesize them. Juchem (2007) indicated that between 70 and 85% of the PUFAs get biohydrogenated in the rumen when cows are fed with unprotected oils. Despite extensive biohydrogenation of unsaturated FA, feeding increasing amounts of ω-6 and ω-3 FA alters tissue composition and influences cellular function and animal performance and reproductive functions (Silvestre *et al.,* 2011). Therefore, it is clear that despite limitations in delivery of specific amounts of polyunsaturated fatty acid for absorption, altering the FA composition of the diet is capable of influencing animal performance. Hence, this experiment was conducted to study effect feeding ω-3 and ω-6 fatty acid rich oil on nutrient digestibility and growth performance of sahiwal heifers.

MATERIALS AND METHODS

The experiment was performed after taking due permission from Institutional Animal Ethics Committee, CPCSEA, New Delhi. The experiment was conducted at cattle farm, Department of Animal Genetics and Breeding, LUVAS, Hisar, using 18 Sahiwal heifers with no overt clinical sickness. The heifers averaged 194±4.16 kg in body weight and 18.33±1.14 months in age. The animals were assigned to 3 groups *viz.* T_1, T_2 and T_3 of 6 animals each following completely randomized design. Animals were dewormed against internal parasites and dipped against ecto-parasites before the start of the experiment following standard protocol. Routine vaccination against Hemorrhagic Septicemia and Foot and Mouth disease was done. All experimental animals were housed in feeding stalls having arrangements for individual feeding and watering. The animals under different dietary treatments were maintained under similar husbandry practices. All the animals were fed isonitrogenous and isoenergetic diets containing weighed quantity of green fodder and concentrate mixture; and wheat straw was fed free of choice. The concentrate mixture fed to heifers in groups T_2 and T_3 was supplemented with soybean oil as ω-6 PUFA source and linseed oil as ω-3 PUFA source, respectively @ 3.5% (3.63 kg per 100 kg). To make the diets isocaloric, palm oil was added in the concentrate mixture of control (T_1). The diets were fed twice daily at 8:00 AM and 4:00 PM. The proximate compositions of feed ingredients, fodder, wheat straw and the different concentrate mixtures were analyzed according to methods of AOAC (2005) and cell wall fractions were determined as per the procedure of Van Soest *et al.* (1991).

Amount of feed offered, residues left and body weight were measured at fortnightly interval for two consecutive days. A digestion trial of 7 days collection period was conducted during which quantitative collection of total feces voided on 24 hourly bases was made to determine the nutrients digestibility of nutrients. Feed cost was calculated according to the price of different feedstuff during the experiment at local market. Data obtained were subjected to statistical analysis as per Snedecor and Cochran (1994) following procedures for Completely Randomized Design (CRD).

RESULTS AND DISCUSSION

Dry matter and crude protein content of T_1, T_2 and T_3 were 93.16, 92.92 and 93.24 percent; and 21.25, 20.98 and 21.14 percent, respectively, indicating that concentrate mixtures were iso-nitrogenous. Ether extract and nitrogen free extract content were 5.32, 5.18 and 5.57; and 56.55, 55.22 and 55.03 percent in concentrate mixture of groups T_1, T_2 and T_3, respectively which revealed that all the concentrate mixtures were isocaloric. Accordingly, TDN intake and TDN content of the diets were also similar. The crude protein content of wheat straw and green fodder were 2.45 and 10.45 percent, respectively. Neutral detergent fibre (NDF) and acid detergent fibre (ADF) contents in concentrate mixture of T_1, T_2 and T_3 were 20.20, 20.86 and 19.63 percent; and 14.30, 14.50 and 14.35, respectively. The NDF and ADF content of wheat straw and green fodder were 75.82 and 50.37 percent; and 43.5 and 33.96 percent, respectively.

Data pertaining to intake and digestibility of nutrients are presented in Table 2. Intake of DM, CP, EE, CF, NFE, NDF and ADF was similar among the groups. The lack of significant effects on intake of these nutrients agrees with results of Muller *et al.* (2004) who showed that ω-3 and ω-6 fatty acids supplementation in crossbred heifers resulted in no differences (P>0.05) in intake of DM, CP, and NDF and ADF. Fiorentini *et al.* (2013) also found no effect of adding soybean oil to the diets of crossbred heifers on nutrient intakes.

In the present investigation PUFA supplementation increased (P<0.05) the organic matter digestibility in T_2, and T_3 groups by 4.01 and 6.20 percent over that of control group. This corroborates well with the earlier reports by Kumar and Thakur (2007). Linseed oil supplementation increased the intake of digestible OM significantly (P<0.05) in T_3 (3160.35g). Linseed as well as soybean oil, both increased the digestible EE intake significantly (P<0.05) which might be due to the numerically higher d igestibility coefficient of EE in PUFA fed heifers. In the same way, Len *et al.* (2016) reported that dietary supplementation of fat sources rich in PUFA has

Table 1. Ingredient (kg/100 kg) and chemical composition (% DM basis) of different concentrate mixtures

Attributes	Concentrate mixtures			Roughages	
	T_1	T_2	T_3	Maize fodder	Wheat straw
Ingredient Composition					
Maize	30	30	30	-	-
Groundnut cake	25	25	25	-	-
Mustard cake	10	10	10	-	-
Wheat bran	32	32	32	-	-
Mineral mixture	2	2	2	-	-
Common salt	1	1	1	-	-
Total, kg	100	100	100	-	-
Palm oil, kg	3.63	-	-	-	-
Soybean oil, kg	-	3.63	-	-	-
Linseed oil, kg	-	-	3.63	-	-
Chemical composition					
DM (%)	93.16	92.92	93.24	25.11	94.43
OM	91.42	91.17	91.03	89.63	90.73
Crude protein	21.25	20.98	21.14	10.45	2.45
Crude fibre	8.30	9.79	9.29	26.40	36.79
Ether extract	5.32	5.18	5.57	4.31	2.87
NFE	56.55	55.22	55.03	48.47	47.62
Total ash	8.58	8.83	8.97	10.37	9.27
NDF	20.20	20.86	19.63	70.69	80.01
ADF	14.30	14.50	14.35	34.92	52.46

positive effects in the improvement of digestibility of EE and OM in Murrah buffalo calves. The CP% and TDN% of the different TMR (Table 3) were not affected due to PUFA addition. Similarly, DCP content was also statistically similar in all the rations. No noteworthy changes were seen in intake of CP, DCP and TDN among the different groups. CP intake varied (P>0.05) from 674.76 g/day in control to 685.40 g/day in T_3. DCP intake was 480.46, 491.81 and 507.44 g/day in T_1, T_2 and T_3, respectively. Dietary PUFA supplementation increased the daily TDN intake from 3.251 kg/day in control to 3..352 kg/d in soybean and 3.491 kg/day in linseed oil fed heifers.

The overall mean values of daily total dry matter intake were 6.01, 6.25 and 6.67 kg in treatment groups T_1 (palm oil), T_2 (soybean oil) and T_3 (linseed oil), respectively (Table 4). The statistical analysis of the data revealed that daily dry matter intake was significantly (P<0.05) higher in heifers fed linseed oil supplemented concentrate mixture as compared to palm oil or soybean oil supplemented groups. The higher value in linseed oil fed group might be due to comparatively more body weight gain in that group. Similar to this study, Childs *et al.* (2008c) also reported higher overall feed intake in fish oil supplemented crossbred heifers as source of ω-3 fatty acids compared to control.

Table 2. Intake and apparent digestibility of nutrients in heifers under different dietary treatment groups

Attribute		Treatments		
		T_1	T_2	T_3
DM	DM Intake, kg/d	5.00±0.03	4.97±0.18	5.14±0.06
	% digestibility	66.58±0.85	67.59±1.74	67.51±2.55
	DDM Intake, kg/d	3.33±0.04	3.38±0.20	3.47±0.14
OM	OM Intake, g/d	4434.84±66.31	4441.13±179.12	4419.25±148.62
	% digestibility	65.38^{b}±0.67	69.39^{a}±1.22	71.58^{a}±0.60
	DOM Intake, g/d	2898.15^{b}±33.04	3071.52^{ab}±75.32	3160.35^{a}±91.20
CP	CP Intake, g/d	674.76±1.27	674.00±6.68	685.40±1.93
	% digestibility	71.20±0.65	72.91±1.31	74.04±1.96
	DCP Intake, g/d	480.46±4.88	491.81±13.23	507.44±13.52
EE	EE Intake, g/d	244.76±5.83	246.32±0.12	248.83±0.62
	% digestibility	71.00±1.88	74.57±0.83	74.85±1.70
	DEE Intake, g/d	173.32^{b}±2.44	183.67^{a}±2.06	186.26^{a}±4.44
CF	CF Intake, g/d	1101.02±15.60	1106.59±53.07	1194.16±4.53
	% digestibility	58.28±2.23	58.53±2.39	57.43±3.15
	DCF Intake, g/dd	641.76±26.31	653.94±56.57	686.33±39.28
NFE	NFE Intake, g/d	2468.22±21.95	2505.14±89.31	2609.70±10.72
	% digestibility	70.41±1.68	71.34±1.28	71.93±2.75
	DNFE Intake, g/d	1738.28±48.66	1792.40±93.52	1878.45±79.31
NDF	NDF Intake, g/d	3059.06±18.63	3121.16±128.03	3107.97±87.51
	% digestibility	60.32±1.93	59.51±2.27	58.93±2.45
	DNDF Intake, g/d	1846.22±65.46	1871.60±145.41	1828.63±82.05
ADF	ADF Intake, g/d	1639.13±26.65	1600.62±70.32	1691.70±11.55
	% digestibility	53.25±2.69	52.58±2.87	51.77±4.34
	DADF Intake, g/d	874.99±54.66	851.15±81.67	877.06±77.09

The mean values bearing different superscripts in a row differ significantly (P<0.05)

Table 3. Mean nutrient intake and nutritive value of experimental ration under different dietary treatments

Attributes		Treatments		
		T_1	T_2	T_3
CP	CP%	11.65±0.02	11.64±0.12	11.84±0.03
	Intake, g/day	674.76±1.27	674.00±6.68	685.40±1.93
DCP	DCP%	8.30±0.08	8.49±0.23	8.76±0.23
	Intake, g/day	480.46±4.88	491.81±13.23	507.44±13.52
TDN	TDN%	55.01±1.53	56.70±3.26	59.49±2.63
	Intake, kg/day	3.251±0.08	3.352±0.16	3.491±0.13

There was no significant effect on DMI per 100 kg body weight by supplementing palm oil or soybean oil or linseed oil in concentrate mixture of Sahiwal heifers. Similar trend was observed when dry matter intake was calculated in terms of dry matter intake per kg metabolic body size, which did not differ significantly

Table 4. Mean dry matter intake, growth performance and economics of feeding different diets (kg/day, DM basis) in experimental heifers

Attributes	Treatments		
	T_1	T_2	T_3
DMI, kg/d	6.01^{b}±0.12	6.25^{b}±0.11	6.67^{a}±0.14
DMI, %BW	2.54±0.02	2.53±0.03	2.54±0.02
DMI, g/kg $W^{0.75}$	98.99±1.19	100.58±1.29	102.53±1.17
Total DMI, kg	901.53^{b}±18.17	937.62^{b}±16.66	999.98^{a}±20.89
Av. roughage intake, kg	3.31^{b}±0.09	3.56^{b}±0.08	3.96^{a}±0.12
Av. CM intake, kg	2.70±0.07	2.69±0.07	2.70±0.07
Total roughage intake, kg	496.28^{b}±14.19	533.42^{b}±12.02	594.39^{a}±17.27
Total CM intake, kg	405.25±10.61	404.20±10.58	405.59±10.62
Initial BW	185.67±9.30	196.17±4.38	201.16±6.79
Final BW	268.33^{b}±7.11	283.00^{b}±6.42	310.33^{a}±7.79
Total gain, kg	82.67^{b}±3.21	86.83^{b}±6.75	109.17^{a}±7.26
ADG, g	551^{b}±21	579^{b}±45	728^{a}±48
FCE	0.09±0.01	0.09±0.01	0.11±0.01
Days taken to 1^{st} estrous	131.33^{a}±10.31	110.17^{ab}±13.79	80.33^{b}±13.66
Pubertal age, days	713.00^{a}±29	665.33^{ab}±26	623.67^{b}±20
DMI till 1^{st} estrous, kg	789.31±61.95	688.54±86.16 (-12.77%)	535.82±91.09 (-32.12%)
Roughage intake till 1^{st} estrous, kg	434.71±34.12	392.19±49.07	318.12±54.08
CM intake till 1^{st} estrous, kg	354.67^{a}±27.83	297.50^{ab}±37.14 (-16.11)	217.00^{b}±36.86 (-38.82)
CM cost, ₹/kg	24.53	25.51	28.13
Cost of CM feeding till 1^{st} estrous, ₹	8698.34±682.68	7587.95±949.47 (-1110.39, -12.77%)	6101.40±1037.26 (-2596.94, -29.86%)

The mean values bearing different superscripts in a row differ significantly ($P<0.05$)

in different treatment groups. The results of the study are in agreement of earlier finding of Dirandeh *et al.* (2014) who reported that average dry matter intake was not affected by dietary inclusion of roasted soybean and linseed as a source of (P<0.05) ω -6 and ω -3 fatty acid, respectively, in the diets of lactating cattle.

Statistical analysis of the data revealed that total body weight gain during experimental period was significantly (P<0.05) higher in heifers fed linseed oil (ω-3 rich oil) supplemented concentrate mixture as compared to soybean oil (ω-6 rich oil) and palm oil supplemented heifers(Table 4). Similar response with respect to average daily gain (ADG) was also noted. The results corroborate with findings of Childs *et al.* (2008) who found that on feeding of graded level of fish oil as source of ω-3 fatty acids significantly increased (P<0.05) the daily body weight gain of crossbred heifers as compared to non-supplemental groups of heifers. Kumar and Thakur (2007) also confirmed that bypass fat supplementation with varying degree of unsaturation increased growth rate in female buffalo calves. The present findings are also in agreement with the report Hill *et al.* (2011) who reported, ADG and efficiency of calves were improved by feeding ω-3 fatty acid rich feed as compared to control diets.

The results of the study revealed that numerical values of FCE improved upon supplementation of ω-3 rich FA oil, however, the difference was statistically non-significant. In earlier report, Garcia *et al.* (2014) revealed that increasing mean intake of linoleic acid from approximately 4.6 to 11.0 g/day during the first 60 day of life increased average daily gain (0.50 *vs.* 0.45 kg/d) without a change in dry matter intake, thus improving feed efficiency.

The cost of feeding during experimental period and up to attainment of first estrous in Sahiwal heifers under different dietary treatments have been depicted in Table 4. The results of the study revealed that daily dry matter intake and total dry matter intake during 150 days of experimental period was significantly (P<0.05) higher in the animals fed concentrate mixture containing ω-3 fatty acid rich oil (T_3) as compared to ω-6 (T_2) and saturated fatty acid (T_1) rich oils. However, the experimental heifers fed soybean oil added concentrate mixture did not show any significant effect of dry matter intake over control group. The mean number of days taken from initiation of experiment to first estrous sign was 131.33, 110.17 and 80.33 days in T_1, T_2 and T_3, respectively, indicating that supplementation of linseed oil has significantly (P<0.05) reduced the days for first sign of heat over soybean and saturated fatty acid oils.

The findings of present investigation revealed that supplementation of ω-3 rich linseed oil in the concentrate mixture of Sahiwal heifers resulted in significant (P<0.05) reduction in the pubertal age of heifers (623.67 days) as compared to ω-6 rich soybean oil supplemented group (665.33 days) and control group (713 days) despite the fact that all the diets were isocalric and iso-nitrogenous. Tran *et al.* (2016) reported that feeding of ω-3 PUFA enriched diet reduced pubertal age and improved both fresh and post-thawing semen quality in male buffalo. The total dry matter intake up to first estrous was reduced by 32.12 and 12.77 percent in T_3 and T_2, respectively, as compared to control (T_1). Similarly, total concentrate mixture intake up to attainment of heifers' pubertal age in ω-3 rich linseed oil and ω-6 rich soybean oil fed group was reduced by 38.82 and 16.11 percent, respectively, over control diet. Hence, the perusal of data on feed cost demonstrated that there was a net saving of ₹ 2596.94 and 1110.39 in Sahiwal heifers by feeding PUFA rich linseed and soybean oil, respectively, from the age 18.33 months to pubertal age.

CONCLUSION

The results of the present study inferred that feeding ω-3 and ω-6 fatty acid rich oil to Sahiwal heifers leads to improved organic matter digestibility and increased intake of digestible ether extract. In addition to that, a diet enriched with source of ω-3 FA such as linseed oil might be a candidate diet to be fed to pre-pubertal heifers to increase the average daily gain; and decrease pubertal age and the cost of feeding.

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