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Effects of feeding flaxseeds and rapeseeds on nutrient intake, nutrient digestibility, feed efficiency and rumen fermentation characteristics in cross bred dairy cattle

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Abstract
Effects of feeding oilseeds on nutrient intake, nutrient digestibility, feed efficiency and rumen fermentation characteristics in fifteen multiparous HF × Kankrej dairy cows were studied subjected to three feeding treatments *viz*., control (T1): TMR without oil seeds; T2: TMR with ground flax seeds @ 4.5 % and T3: TMR with ground rapeseed @ 4.5%. The experimental TMR were offered along with 15 kg green cereal fodder as per the guidelines of ICAR (2013) standards. The results revealed the DMI, CPI and TDNI were similar and within the normal range in all the groups with non-significant differences among the groups. While DCPI was found ($P<0.05$) higher by 13% in T3 group. The digestibility of DM, CP, NFE, CF, NDF, ADF, HC and cellulose did not reveal any change on account of dietary treatments. Only EED was ($P<0.05$) observed higher in T3 group by 11.56%. Similarly, the feed efficiency was also remained at par among the treatments. Ruminal pH, total nitrogen (N), soluble N, ammoniacal nitrogen, total volatile fatty acids and microbial protein synthesis were not affected by dietary treatments. However non protein nitrogen (NPN) was found ($P<0.05$) higher in T2 group, while T1 remained at par to both T2 and T3. It was concluded that oilseeds inclusion in diets of dairy cattle did not give any adverse effect on nutrient intake, its digestibility, feed efficiency and rumen fermentation.

Keywords: Oilseeds, nutrient digestibility, nutrient intake, rumen fermentation, dairy cows

Introduction
Food is one of the most important and modifiable lifestyle determinants of human health. In present scenario the functional and nutraceutical food are more preferred and consumed by the health-conscious people. Milk is merely a complete food and consumed by almost everyone every day. Composition of milk revealed that it has almost double the saturated fatty acids and half the unsaturated fatty acids compared to the ideal milk composition for human beings [1].
Omega-3 fatty acids are beneficial as it has anti-inflammatory effect while omega-6 has pro-inflammatory effect. Several researches have shown health benefits of omega-3 fatty acids to humans including a decrease in the incidence of cancer, cardiovascular diseases, hypertension, and arthritis and an improvement of visual acuity [2].
Oil seeds are one of the main sources of unsaturated fatty acids among which flaxseeds and rapeseeds are rich source of omega-3. Feeding oilseeds to the dairy animals is one of the way of dietary intervention to manipulate the saturated to unsaturated fatty acid ratio in milk fat. Flaxseed (*Linum usitatissimum)* contains about 20% protein and 40% oil [3, 4] which makes it an interesting feed ingredient for inclusion in lactating dairy cows' rations as a source of both. Flaxseed has high linolenic acid constituting approximately 55% of total fatty acids [4, 5]. Rapeseed (*Brassica napus*) contains 41– 43 % oil, of which about 12 % is ALA. Rapeseeds are considered as a good source of cis-9 18:1 and cis-13 22:1 as well as protein (20–25 %). Rapeseed is a good source of monounsaturated (22-60 %) fatty acid and poly unsaturated fatty (14%) acids [6]. With an objective to improve the milk fatty acid profile of dairy cattle by dietary interventions, it was required to study the effects of supplementation of two oilseeds on nutrient intake, nutrient digestibility and feed efficiency and rumen fermentation characteristics in HF × Kankrej (50:50) cross bred dairy cattle.

Materials and methods
Animal, Housing and Feeding
Fifteen multiparous HF × Kankrej (50:50) dairy cows were selected to assigned one of the three dietary treatments in a complete randomised design based on previous lactation.

Each treatment group was comprised of five animals and they were fed with the experimental diets. The composition of the experimental diets is given in Table 1. The experimental period was of 120 days excluding 15 days from parturition. The experimental TMR were offered along with 15 kg green cereal fodder as per the guidelines of ICAR (2013) standards. All the experimental animals were kept iso-managerial and kept in well ventilated hygienic condition and they were provided with feed and fodders as per treatments. Throughout the experimental period, the animals were maintained in an open asbestos sheeted shed with cemented floor, having arrangement for individual feeding. The three feeding treatments were., control (T1): TMR without oil seeds; T2: TMR with ground flax seeds @ 4.5 % and T3: TMR with ground rapeseed @ 4.5%. Green fodder was offered to animals just after tying to shed around 11:00 A.M. and measured quantity of TMR were offered to the animals in two parts, during afternoon at around 02:30 P.M, and remaining proportion was offered after milking at around 05:30 P.M. During both the morning and evening milking, animals were offered with 1 kg TMR. Targeted daily intake of linseed and rapeseed was 0.5 kg/cow. Diets were designed to be isonitrogenous.

Table 1: Ingredients and chemical composition of the diets

Ingredients	Composition of experimental rations (%)		
	Control (T1)	TMR with flaxseed (T2)	TMR with rapeseed (T3)
Cereal straw	45	45	45
Soyabean meal	17	15.5	15.5
Maize	13	11.5	11.5
Rice	13	11.5	11.5
Flaxseed	-	4.5	-
Rapeseed	-	-	4.5
Molasses	10	10	10
Mineral mixture	2	2	2
Chemical composition (% on DM basis)			
Crude protein (CP)	12.40	12.56	12.55
Ether extract (EE)	2.67	2.99	4.43
Crude fibre (CF)	29.22	24.94	22.35
Total ash (TA)	10.74	10.96	10.53
Nitrogen free extract (NFE)	44.97	48.53	50.12
Acid insoluble ash(AIA)	5.70	5.59	5.27
Neutral detergent fibre (NDF)	58.44	55.00	55.40
Acid detergent fibre (ADF)	30.07	31.76	31.89
Cellulose (C)	23.82	22.73	22.55
Hemicellulose (HC)	28.37	23.24	23.51
Acid detergent lignin (ADL)	14.30	10.78	9.56

Nutrient intake and apparent nutrient digestibility

Daily intake and refusal of green and TMR was recorded for individual animals and TMR were sampled weekly. A digestion trial was conducted in mid of experimental period with 7 days collection period to determine the digestibility of nutrients. Proximate analysis of feed materials was carried out as per the methodology of AOAC [7] and the fraction of cell wall constituents such as NDF, ADF and cellulose were estimated as per the procedure of Van Soest [8].

Rumen studies

Collection of rumen liquor

Rumen liquor was collected at 3 h pre and post feeding through a stomach tube against negative pressure created by a suction pump adult crossbred experimental animal. The collected rumen liquor was strained through four layered muslin cloth and was referred as Strained Rumen Liquor (SRL). The SRL was brought to the laboratory in a pre-warmed (39±1°C) thermos flask.

Processing of rumen liquor

About 150 ml of rumen liquor was collected from each experimental animal at 0, 3 and 6 h post feeding through a stomach tube against negative pressure created by a suction pump using method developed by Lane [9]. The rumen liquor was immediately brought to the laboratory and strained through four layered muslin cloth.

pH determination

The pH of SRL was determined immediately after collection using portable digital pH meter. After pH determination, one ml of saturated $HgCl_2$ solution was added to each collected sample to kill the microbes and to stop metabolic activity.

Nitrogen fractions and total volatile fatty acids

The samples of SRL were analyzed for ammonia-N using the method given by [10] and total-N by Kjeldahl's method. Soluble-N in supernatant of SRL after centrifuging was estimated by Kjeldahl's method and non-protein-nitrogen estimated by Trichloroacetic acid precipitation of SRL and estimating the N content of supernatant by Kjeldahl's method. The concentration of total VFA was determined in SRL by the steam distillation method given by [11] using Markham micro-distillation apparatus.

Statistical analysis

Observations of various parameters were recorded during experimental period and tabulated data were statistically analysed by Completely Randomized Design (Factorial) as described by wasp software. Some of the data were also analysed by one-way ANOVA using SPSS software 20.00 version.

Results and Discussion

The ingredients and chemical composition of three diets are presented in Table: 1. The chemical composition of the TMR

was generally similar among diets, except for EE, which was lowest for the control diet. The nutrient intake and digestibility are presented in Table: 2.

The average daily DMI was 12.88, 12.82 and 12.48 kg in T1, T2 and T3, respectively. It was similar in all the groups with non-significant differences. The dry matter intake was observed within the normal range in all the groups which indicated no adverse effect of flaxseed and rapeseed on palatability and feed intake of animals. Similar to present findings, no-significant effect of feeding flaxseeds to dairy cattle in different forms has been reported by various authors; [4, 12-16]. However, higher DMI was also reported by [3] while [17, 18] observed lowered DMI by feeding different forms of flaxseed in the ruminant diet. Similarly many studies [19–22] agrees with the results of current study for rapeseed. In contrast to this [23] and [24] reported higher and lower DMI in rapeseed fed animals respectively.

The average CPI in T1, T2 and T3 groups were 1.50, 1.51 and 1.47 kg/head respectively. The CPI was more or less similar in all the groups which agrees with other studies [25, 26] and [24] for flaxseed and rapeseed feeding in dairy cows. The average DCPI was 0.77, 0.81 and 0.87 kg/head in T1, T2 and T3 groups, respectively. The DCPI in T3 significantly increased by 13% while in T2 it was similar in T1 and T2. Higher DCPI was due to higher CP digestibility in T3.

The average TDNI in T1, T2 and T3 treatments was 7.11, 7.04 and 7.30 kg/head, respectively. The TDNI in T1 and T2 remained similar while in T3 group it was increased by 2.67%. Though the difference was non-significant.

Table 2: Nutrient intake, digestibility and feed efficiency during the experiment

Item	T1	T2	T3	Sem	P value
Nutrient intake kg/d					
DMI kg/d	12.88	12.82	12.48	0.08	>0.05
CPI kg/d	1.50	1.51	1.47	0.31	>0.05
DCPI kg/day	0.77^{b}	0.81^{ab}	0.87^{a}	0.02	<0.05
TDNI kg/day	7.11	7.04	7.30	0.19	>0.05
Digestibility % of item					
DMD	54.15	53.70	57.70	1.98	>0.05
EED	70.70^{b}	67.60^{b}	78.87^{a}	2.49	<0.05
CPD	51.37	53.82	59.31	3.54	>0.05
NFED	68.78	70.63	74.66	1.74	>0.05
CFD	48.66	42.43	47.47	2.78	>0.05
NDFD	49.47	47.24	52.24	2.28	>0.05
ADFD	42.88	46.16	51.32	2.83	>0.05
CD	56.22	53.56	62.55	2.93	>0.05
HCD	57.05	50.82	53.46	2.20	>0.05
Feed Efficiency					
Milk production kg/ kg DMI	0.79	0.77	0.73	0.08	>0.05
Milk production kg/ kg CPI	6.84	6.56	6.14	0.69	>0.05
Milk production kg/ kg DCPI	13.31	12.20	10.36	1.29	>0.05
Milk production kg/ kg TDNI	1.44	1.41	1.24	0.14	>0.05

^{abc} Means within a row with no common superscript differ, (P=0.05).

The average DM digestibility coefficients (%) under T1, T2 and T3 groups was 54.15, 53.70 and 57.7% respectively. The DM digestibility of T1 and T2 groups was all most similar while in T3 group it increased by 6.65% as compared to control group, though the difference was non-significant.

The average CP digestibility coefficients under T1, T2 and T3 groups were 51.37, 53.82 and 59.31%, respectively. The CPD increased by 4.77 and 15.46% in T2 and T3, respectively. Though the differences were non-significant among the groups. The present study agrees with findings of many researchers in flaxseed [4, 12, 27-29] and rapeseed [30] fed dairy cows in different forms. However, [31] observed 5.5% higher CPD in extruded flaxseed fed dairy cows as compared to untreated flaxseed fed animals.

The average EE digestibility coefficients under T1, T2 and T3 groups were 70.7, 67.6 and 78.87%, respectively. The EED was similar in T1 and T2 groups, however it was significantly increased by 11.56% in T3 group. Similar, result was found by [12, 28, 32] in flaxseed fed dairy cows. While [29] found significantly increased EED by 9% in flaxseed oil (@2% on DM basis) fed animals than that of control group. However, [33] reported at par EED in cows fed with or without canola which does not agree with the current study.

The average NFED coefficients in T1, T2 and T3 groups was 68.78, 70.63 and 74.66%, respectively. The NFED in T2 and T3 group was increased by 2.69 and 8.54% as compare to T1. However, the difference was non-significant.

The average CF digestibility coefficients under T1, T2 and T3 groups were 48.66, 42.43 and 47.47%, respectively. In the present study the CFD was decreased in T2 and T3 group by 12.8 and 2.44%, respectively, though the difference remained non-significant among the treatments. This indicates oilseeds inclusion in the diet does not affect the CFD. The present study agrees with [34] observed non-significant changes on CF digestibility in dairy cows fed with canola oil.

The average NDF digestibility coefficients under T1, T2 and T3 groups was 49.47, 47.24 and 52.24%, respectively. The NDFD increased by 5.59% in T3, though the difference was non-significant. In flaxseed fed animals similar results were obtained by [12, 28] while for rapeseed [24, 30, 33] obtained similar results. In contrast to this study, [29] observed 20% more NDFD in flaxseed oil fed animals as compared to control.

The average ADF digestibility coefficients under T1, T2 and T3 groups were 42.88, 46.16 and 51.32% respectively. The ADFD increased by 7.65, and 19.68% in T2 and T3 group, respectively, though the difference was non-significant. Similar results were found by [12, 29]. The average HC digestibility coefficients under T1, T2 and T3 groups were 57.05, 50.82 and 53.46%, respectively. The HCD decreased by 6.23, 6.29% in T2 and T3 group respectively. Though the difference was non-significant.

The average cellulose digestibility coefficients under T1, T2 and T3 groups were 56.22, 53.56 and 62.55% respectively. The CD was increased and decreased by 11.26, and 4.73% in T3 and T2 group respectively, though the difference was non-significant.

The average milk production kg/ kg of DMI, CPI, DCPI and DCPI are mentioned in Table: 2. The all parameters were remained non-significant among all the groups which shows no adverse effect of feed-on-feed efficiency. The rumen fermentation characteristics has been mentioned in Table:3.

Table 3: Ruminal fermentation parameters of dairy cows fed with oilseeds

Item	T1	T2	T3	Sem	P value
pH	7.30	7.26	7.47	0.12	P>0.05
Total nitrogen (mg/dl)	108.00	93.00	73.90	9.48	P>0.05
Soluble nitrogen (mg/dl)	16.89	18.39	18.39	0.98	P>0.05
Ammoniacal nitrogen (mg/dl)	18.39	18.95	17.09	2.01	P>0.05
NPN (mg/dl)	42.37^{ab}	46.66^{a}	36.03^{b}	2.39	P<0.05
Total volatile fatty acids (mmol/dl)	8.03	8.72	4.87	1.97	P>0.05
Microbial nitrogen (g/day)	69.13	72.04	83.84	6.20	P>0.05

^{abc} Means within a row with no common superscript differ, (P=0.05).

The ruminal pH under T1, T2 and T3 were 7.30, 7.26 and 7.47 respectively which was at par in all the groups. This shows no adverse effect was found on ruminal environment due to incorporation of oilseeds in treatment groups. The similar type of results were found by [28, 32, 35, 36] in flaxseed fed animals while [37] found increased rumen pH in flaxseed fed animals. In contrast to this [33] found decreased pH in canola seed fed animals as compare to control diet.

The total nitrogen was 108.00, 93.00 and 73.90 mg/dl in T1, T2 and T3 groups, respectively. The total nitrogen was decreased by 13.88 and 31.57% in T2 and T3, respectively. Though the difference was non-significant. The soluble nitrogen in T1, T2 and T3 was 16.89, 18.39 and 18.39 mg/dl, respectively. However, it was increased by 8.88% in both T2 and T3 groups as compare to T1, the difference remained non-significant.

The ammoniacal nitrogen was 18.39, 18.95 and 17.09 mg/dl in T1, T2 and T3 groups, respectively. It was decreased by 7.01% in T3 group as compare to T1 while in T2 it was remained at par to T1. Though the difference was non-significant. Similar kind of results were found by [28, 36] for flaxseed fed animals in different forms.

The NPN was found 42.37, 46.66 and 36.03 mg/dl in T1, T2 and T3 respectively which was significantly increased in T2 group as compare to T3 group. However, control group remained at par to T2 and T3.

The total volatile fatty acids in T1, T2 and T3 were 8.03, 8.72 and 4.87 mmol/dl respectively. [28, 35-37] and [33] also found similar type of results for flaxseeds and rapeseeds fed animals in different forms respectively.

The microbial protein synthesis was remained unaffected by oilseeds incorporation in all three groups. It was found 69.13, 72.04 and 83.84 (g/day) in T1, T2 and T3, respectively. This indicates inclusion of oilseeds in dairy animal diets did not give any adverse effect on ruminal ecosystem.

Conclusions

The result revealed that TMR containing 4.5% flaxseed (T2) and 4.5% rapeseed (T3) has no adverse effect on palatability of the feed as voluntary nutrient intake remained similar. Incorporation of oilseeds did not confer any negative changes in the digestibility of the crude fibre fraction. Rapeseed containing diet improved the digestibility of ether extract. Feed efficiency was also found to be at par among all the experimental treatments. The ruminal fermentation characteristics were also remained similar in all three groups which indicates inclusion of oilseeds did not affect the rumen environment and its fermentation pattern. However only non-protein nitrogen was found significantly higher in T2 group.

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