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Effect of Herbal Feed Additives and Sulfate Supplementation on Growth Performance and Nutrient Utilization of Cattle Calves

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ABSTRACT

Background: Fibre is digested in the foregut of all ruminant animals by the microbes present. Digestibility of fibres of crop residues is low along with significant loss of feed energy as methane. Sulfate is reduced in the rumen by sulfate-reducing bacteria (SRB) and stimulates microbial growth with an increase in rumen microbial protein supply to the host animal. Essential oils (EOs) are among the most important plant-derived products, frequently responsible for the health-promoting qualities of various species. Both essential oils from fennel and clove exhibited pronounced and varying degree of growth inhibition against fungal and bacterial pathogens. Microbial digestion in the rumen can be improved by using these feed additives. Therefore, in the current study a combination of herbal feed additive and sulfate test in *in vivo* trial for their effect on nutrient utilization and performance of calves.

Methods: Twenty four growing cattle calves (12 male Haryana, 8 male Sahiwal, 4 female Sahiwal calves) were distributed into four groups with six animals each in a randomized block design. Control (C) group was not supplemented with any extra things other than the present in the basal diet, T1 group was supplemented with herbal feed additive (Fennel + CLO; @ 1.0% of DMI), T2 group was supplemented with sulfate @ 0.075% of DMI and group T3 were supplemented with diet containing herbal feed additive (Fennel + CLO; @ 1.0% of DMI) and sulfate @ 0.075% of DMI. The basal diet offered to experimental groups contains 40% concentrate and 60% wheat straw. Body weight and dry matter intake were recorded fortnightly.

Result: Overall body weight, DMI (kg/day), TDN intake (g/kg $W^{0.75}$) and DCP intake (g/kg W) were found to be similar in the treatment as well as control group. Nutrient digestibility and digestible nutrient intake were not impacted by supplementation of either herbal feed additive and/or sulfate supplementation. ADGs were similar in all groups. FCR was not significantly different between the control and treatment group**s.** It may be concluded that supplementation of herbal feed additive (Fennel + CLO; @ 1.0% of DMI) may be used as an alternate electron acceptor and rumen modifier in high roughage-based ration as it does not have any harmful effect on growth and nutrient utilization on cattle calves.

Key words: Feed additive, Growth performance, Nutrient utilization, Sulfate.

INTRODUCTION

India possesses one of the largest populations of ruminants in the world which have an important role in the sustainability of livestock farmers and many cases are the major source of income for small and marginal farmers. These farmers rely on subsistence farming, with few or no inputs, where poor quality forages are usually available and provide the sole source of nutrition to the animal. Currently, the livestock sector's main challenge is providing essential nutrients through feed supplementation to promote health and production to the optimum genetic potential level. Digestibility of roughages like straws is very poor, so these plant secondary metabolites (PSM) like tannin, saponin and essential oils can improve the digestibility. Essential oils (EOs) are among the most economically relevant plant-derived products, being frequently responsible for several species' health-promoting properties (Mushtaq *et al.,* 2018). Microbial digestion in the rumen can be improved by using feed additives. Some additives could drive the fermentation reactions toward the formation of more efficient products by acting on the balance of the microbial population. Antibiotics have been widely used in the past as microbiota regulators. However, their use as feed additives has been

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banned in the EU since 2006. In this context, EOs could offer some new opportunities to replace antibiotics. EOs present a high affinity for the membrane cell of bacteria due to their hydrophobic nature and their affinity for lipids which can disrupt the cytoplasmic membrane either directly or by damaging the membrane proteins, provoking increased membrane permeability, conformational changes, leakage of cytoplasmic components, interfering with bacterial growth and activity, causing changes in the rumen population and fermentation profile. They are volatile constituents of terpenoid or non-terpenoid origin which impair the energy metabolism of archaea and have shown reductions of up to 26% in enteric methane emissions in ruminants (Patra and saxena, 2009). Fennel seed also has antimicrobial activity (Bhaisare *et al.,* 2015). Due to the adverse effects on feed digestion and fermentation at high doses, a single EO may not effectively and practically mitigate methane emissions from ruminants unless used at low doses in combination with other antimethanogenic compounds. Therefore, it seems that sulphate (efficient electron acceptors) along with plant secondary metabolites can act as rumen modifiers to mitigate methane production and improved nutrients utilization.

Therefore, in the present study, a combination of herbal feed additive and sulfate is proposed to test in *in vivo* trial for their effect on nutrient utilization and the performance of calves.

MATERIALS AND METHODS

Selection of experimental animals, dietary treatment and experimental design

Twenty four growing cattle calves (12 male Hariana calves, 8 male Sahiwal calves and 4 female Sahiwal calves) were selected from the herd maintained at Livestock Farm Complex (LFC), DUVASU Mathura (Uttar Pradesh) India. Animal care procedures were approved (approval number IAEC/22/2/18) and conducted under the established standard of the Institutional Animal Ethics Committee (IAEC), constituted as per article number 13 of the Committee for Control and Supervision of Experiments on Animals (CPCSEA) rules laid down by the Government of India. Calves with were divided into four groups (six animals each) in a randomized block design. The four groups of calves with were allotted to different treatments. Control (C) group was not supplemented with any extra things other than the present in the basal diet, T1 group were supplemented with herbal feed additive (Fennel + CLO; @ 1.0% of DMI), T2 groups were supplemented with sulfate @ 0.075% of DMI and group T3 were supplemented with diet containing herbal feed additive (Fennel + CLO; @ 1.0% of DMI) and sulfate @ 0.075% of DMI. Feeding was done as per ICAR (2013) to meet the requirement for an average daily gain (ADG) of 500 grams. The basal diet offered to experimental groups contains 40% concentrate and 60% wheat straw. The experiment was continued for 120 days where all the calves were managed under similar conditions.

A digestion trial of six days was conducted on all the experimental animals after 120 days of experimental feeding. During the digestion trial, daily faeces voided, feed offered and residues left were recorded. 24h faecal output of each animal was quantified and 25% of the voided faecal sample was dried in forced draught hot air oven at 60°C. The dried faecal sample of individual animals of each day of six day collection period was bulked, sampled and ground in a hammer mill to pass through 2 mm sieve. The ground sample were stored in airtight bottles until required for chemical analysis. The growth trial lasted for 120 days during which feed intake was recorded daily for each calf. Calves' body weight was recorded for two consecutive days fortnightly immediately before offering feed and water and these values were used to determine body weight gain. The feed offered, residues left and feces voided were analyzed for DM by drying at 100°C for 24 h, OM by ashing at 550°C for 3 h and CP by Kjeldahl technique, NDF was measured by the methodology of Van Soest *et al.* (1991). Ingredients and chemical composition of the basal diet fed during the experimental period are analyzed by AOAC (2005) and presented in Table 1 and 2.

All statistical analyses were performed as per standard method by using SPSS computer package (SPSS Version 20.0, SPSS Inc, Chicago, USA). The data obtained were statistically analyzed by using one-way ANOVA procedures. Tukey's Honest Significant Difference test was used to evaluate significant differences between means of treatments. The difference between means was declared significant at $P<0.05$.

RESULTS AND DISCUSSION

Growth performance

The data of Body weight change and FCR during 120 days experimental trial on growth performance are presented in Table 3 and 4. Average BW at starting of the study in (C), T1, T2 and T3 groups were 80.3, 80.3, 81.8 and 80.3 kg and at the end of the experiment were 129, 127, 129 and 127 kg respectively. The overall BW (kg) was found to be similar in all the experimental groups. Statistical analysis of data

Table 1: Nutrient composition (% or as mentioned) of diet fed during the experimental period.

Attribute (%)	Concentrate	Wheat straw
Dry matter	90	92
Organic matter	84.9	80.7
Crude protein	20.2	3.2
Ether extract	6.24	1.31
Total ash	9.25	13.25
Crude fiber	16.12	38.9
Nitrogen free extract	48.45	43.25
Neutral detergent fiber	33.15	82.25
Ca %	1.43	0.38
P %	0.39	0.17

showed that variation between the groups for mean BW change was not significant (P>0.05). The feed conversion ratio of calves at different fortnight is presented in Table 4. The FCR ranged from 6.12 to 8.81, 6.12 to 9.48, 5.70 to 8.10 and 5.48 to 8.74 in C, T1, T2 and T3 groups, respectively. FCR did not differ significantly (P>0.05) between the groups. Similar to our findings, Kumar (2017) reported no effects on body weight gain in buffalo calves fed a blend of essential oil rich poplar (*Populus deltoids*) and eucalyptus (*Eucalyptus citriodora*) leaves. There was no difference in the growth performance of steers among the treatments on supplementation of 0.1% allicin (Ahn *et al.,* 2024). Bampidis *et al.* (2005) observed no change in Average Daily Gain (ADG) and feed efficiency when grow lambs were fed diets supplemented with oregano leaves (*Origanum vulgare* L.) providing 144 or 288 mg of oregano oil (850 mg of carvacral) per kilogram of diet DM.

Nutrient utilization

Data of apparent nutrient digestibility across six days digestion trial is presented in Table 5 and 6. Statistical analysis of data revealed that variation between the groups for dry matter intake (kg/day) of all the groups during different fortnights of experimental feeding was not significantly different (P>0.05). DMI of control (C), T1, T2 and T3 was 3.12, 3.08, 3.18 and 3.08 kg/d, respectively. The overall DMI (kg/100 kg BW) was also found similar in all the experimental groups. The overall BW (kg) was found similar in all the experimental groups. The mean average daily gain were 410, 413, 416 and 410 g/day in (C), T1, T2 and T3 groups, respectively. The average daily weight gain of all the fortnight were not differ significantly (P>0.05) between groups. FCR of all the fortnight were approximately similar (P>0.05) between groups. The statistical analysis of the data on DM and OM digestibility per cent shows that the

Table 2: Chemical composition (% DM basis) of diet fed to calves during feeding trial.

Ingredients			Dietary group			
			C	T1	T2	T3
Wheat straw			60	60	60	60
	Ingredients	(% DM)				
	Maize grain	15				
	Barley grain	15				
Concentrate	Wheat bran	33	40	40	40	40
	Mustard cake	35				
Mineral mixture (17% Ca)			+	+	+	+

Table 3: Effect of herbal feed additives and sulfate supplementation on fortnight body weight (kg) change.

Fortnight	Control	T1	T2	T3	SEM	P value
0	80.37±20.99	80.36±22.58	81.83±17.76	80.66±13.45	8.86	1.00
1st	83.33±20.67	85.72±22.59	89.00±17.91	88.65±13.36	8.83	0.996
2nd	87.93±20.78	90.10±22.17	92.88±17.91	93.76±13.42	8.80	0.996
3rd	93.83±21.09	94.90±21.93	97.85±17.70	98.34±13.58	8.71	0.998
4th	100.9±21.68	100.6±21.99	104.5±17.66	104.1±13.41	8.81	0.998
5th	107.5±21.68	106.7±22.32	109.4±17.72	110.0±13.43	8.89	0.996
6th	114.6±21.99	112.7±22.40	116.1±17.43	116.7±13.48	8.82	0.996
7th	121.8±21.91	119.9±22.49	123.1±17.43	123.1±13.52	8.94	0.999
8th	129.1±21.46	127.9±22.51	129.3±17.73	127.5±13.53	8.92	0.996
Average	102.1	102.1	104.9	104.7	3.07	0.979

Table 4: Effect of herbal feed additives and sulfate supplementation on FCR.

Fortnight	Control	T1	T2	T3	SEM	P value
1st	6.34±1.81	6.12±0.69	5.70±0.59	5.48±0.71	0.518	0.691
2nd	6.12±0.76	9.48±3.30	6.45±0.88	6.54±0.97	0.908	0.414
3rd	6.95±0.86	7.53±1.68	7.63±1.31	6.91±0.61	0.558	0.958
4th	6.27±0.75	6.99±0.69	6.76±0.97	8.74±1.29	0.487	0.315
5th	7.78±1.12	6.52±0.61	7.24±0.90	7.62±0.97	0.440	0.772
6th	7.21±1.00	8.44±1.14	8.10±1.70	7.37±0.76	0.570	0.869
7th	8.11±1.33	7.33±0.92	7.91±1.13	7.06±0.64	0.492	0.880
8th	8.81±2.26	6.21±0.84	6.97±0.85	6.06±1.44	0.723	0.540
Average	7.20	7.31	7.15	6.97	0.215	0.995

Table 5: Effect of herbal feed additives and sulfate supplementation on nutrients digestibility percentage coefficient.

Attributes	Control	T1	T2	T3	SEM	P value
Initial BW (kg)	126±21.71	125±22.48	126±17.54	124±13.38	8.90	1.000
Final BW (kg)	129±21.46	127±22.51	129±17.73	127±13.53	8.92	0.994
ADG (g/day)	410±8.30	413±8.82	416±11.65	410±12.04	4.77	0.971
DM	57.3±1.00	57.5±1.05	58.4±0.96	57.2±1.04	0.48	0.818
OM	60.4±0.65	60.4±1.04	62.3±1.83	58.6±1.49	0.68	0.320
CP	66.4±1.58	62.4±2.17	64.1±2.04	69.0±0.77	0.97	0.080
CF	50.9±1.63	51.5±1.09	50.3±1.46	50.0±1.35	0.66	0.866
EE	77.0±0.58	77.8±0.46	78.9±0.65	78.9±0.43	0.30	0.070
NFE	60.1±0.91	60.9±0.93	60.2±1.21	61.4±0.81	0.46	0.745
NDF	58.2±1.08	58.3±1.21	58.5±1.23	59.0±0.83	0.51	0.949

C: Control animals having no feed additives supplementation; T1: Treatment Animals supplemented with herbal feed additives; T2: animals supplemented with sulfate; T3: Animals supplemented with herbal feed additives and sulfate; P>0.05: Non-significant

Table 6: Effect of herbal feed additives and sulfate supplementation on nutrient intake during digestion trial.

Attributes	Control	T1	T2	T3	SEM	P value
DMI (Kg/Day)	3.12±0.41	3.08±0.45	3.18±0.34	3.08±0.30	0.173	0.997
DMI (Kg/100kg Body wt)	2.39±0.10	2.37±0.09	2.43±0.10	2.33±0.68	0.043	0.902
DMI (G/Kg W0.75)	79.99±0.41	79.3±0.48	81.6±1.01	78.8±2.53	0.686	0.534
CP intake g/day	257±35.68	259±32.83	274±30.79	278±29.35	14.9	0.954
CPI g/100kg BW	196±9.48	202±10.20	208±8.01	209±5.19	4.06	0.665
CP g/kg W0.75	6.57±0.22	6.72±0.14	7.00±0.17	7.09±0.25	0.104	0.278
DCPI g/day	171±24.66	159±14.17	174±15.51	192±22.03	9.40	0.687
DCPI g/100kg BW	130±7.48	127±9.93	134±8.75	144±3.43	3.88	0.437
DCP g/Kg W0.75)	3.60±0.52	3.59±0.55	3.66±0.55	3.79±0.26	0.223	0.989
TDN (Kg/day)	1.61±0.22	1.57±0.21	1.62±0.18	1.62±0.19	0.091	0.998
TDN/100kgBW	1.23±0.59	1.22±0.59	1.23±0.47	1.21±0.45	0.024	0.996
TDN g/Kg W0.75	41.3±0.85	40.9±0.75	41.5±0.64	41.2±2.02	0.553	0.989

C: Control animals having no feed additives supplementation; T1: Treatment Animals supplemented with herbal feed additives; T2: animals supplemented with sulfate; T3: Animals supplemented with herbal feed additives and sulfate; P>0.05: Non-significant.

differences between the groups are not statistically significant (P>0.05). CP digestibility in calves from four different groups averaged 66.4, 62.4, 64.1 and 69.0 per cent, respectively and nutrients digestibility (CP, CF, EE and NDF) were found similar (P>0.05) among all four groups as mentioned. The experimental calves fed with herbal feed additive and sulfate did not have any significant effect on the DMI and ADG compared to calves in the control group. Digestibility of DM, CP, EE, CF, NFE and NDF were not impacted by supplementation of herbal feed additive and/or sulfate and digestible nutrient intake was found similar in all the experimental groups. Total DMI (kg/day), CP intake (g/day), DCP intake (g/day), TDN intake (g/day) were found similar (P>0.05) in all four, control (C1), T1, T2 and T3 groups as mentioned in table 6. In accordance with our findings, Lakhani *et al.* (2019) also reported no effect on nutrient utilization and digestibility by feeding rumen modifiers and sodium sulfate. Similarily, Wang *et al.* (2025) also reported no effect on nutrient utilization and digestibility by feeding essential oil. Some researchers have demonstrated improvements in performance with herbal feed additives and sulfate supplementation, Souza *et al.* (2019) supplementated diet with clove and rosemary essential oils and protected oils (eugenol, thymol and vanillin) 4gm/animal/day in Nellore heifers and found there is improved average daily gain, dry matter intake, feed efficiency and ingestive behavior activities. Saeedi *et al.* (2017) observed supplementation of fennel seed powder @ 0.4% to the starter diet of Holstein dairy calves improved the feed intake, feed efficiency and growth performance and allowed the calves to be weaned at an early age.

The present study result has no effect on nutrient utilization and growth performance, it may be due to the lower doses of herbs and sulfate in the treatment groups.

CONCLUSION

From this study, it can be concluded that Dietary supplementation of herbal feed additive (Fennel + CLO; @ 1.0% of DMI) and sulfate @ 0.075% of DMI have no significant (P>0.05) effect on BW, ADG, FCR, intake and digestibility of nutrients, indicating it may be used as

alternate electron acceptor and rumen modifier in high roughage-based ration as it does not have harmful effect on growth and nutrient utilization on cattle calves.

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Disclaimers

The views and conclusions expressed in this article are solely those of the authors and do not necessarily represent the views of their affiliated institutions. The authors are responsible for the accuracy and completeness of the information provided, but do not accept any liability for any direct or indirect losses resulting from the use of this content.

Informed consent

All animal procedures for experiments were approved by the Committee of Experimental Animal Care and handling techniques were approved by the University of Animal Care Committee.

Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this article. No funding or sponsorship influenced the design of the study, data collection, analysis, decision to publish, or preparation of the manuscript.

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