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Influence of plant secondary metabolites supplementation on nutrient digestibility in cross-bred dairy cattle

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Abstract
The experiment was conducted to study the effect of plant secondary metabolites supplementation on nutrient intake and nutrient digestibility in cross-bred dairy cattle. Eighteen, lactating cross bred Vrindavani cattle (30 to 60 days postpartum) were selected (based on parity and milk yield) and divided into 3 groups with each group containing 6 animals. Control group was fed with basal diet (concentrate according to milk yield and green fodder *ad libitum*). In T_1 group, basal diet was supplemented with a combination of ajwain oil and bahera. In T_2 group basal diet was supplemented with a combination of ajwain oil and bahera at double the dose of T_1. Nutrient intake, as well as nutrient digestibility (CP, ADF, EE) was similar in all three experimental groups whereas NDF digestibility was significantly (p>0.05) higher T_3 group than control and T_1. DCP and TDN intake in terms of kg/day and g/kg$W^{0.75}$ was comparable among groups. In conclusion, dietary supplementation of plant secondary metabolites did not affect nutrient intake nutrient digestibility in cross-bred dairy cattle except NDF digestibility which was improved.

Keywords: Plant secondary metabolites, nutrient digestibility, dairy cattle

Introduction
Dairy cattle production is a critical component of the global livestock industry, providing a significant source of milk and dairy products for human consumption. Maximizing the efficiency of nutrient utilization in dairy cattle is essential to enhance milk production, improve animal health and reduce production costs. One promising avenue for achieving these goals is the supplementation of plant secondary metabolites (PSMs) in the diet. In recent years, much research has focused on evaluating the potential use of plant secondary metabolites alternatives to feed antibiotics to improve feed efficiency in ruminants (Benchaar *et al*., 2008) [4].

PSMs are diverse groups of chemical substances produced by plants but not engaged in their core metabolic activities of growth and reproduction. Bioactive compounds of PSM have antibacterial properties and are intended to defend the host plant from invading particles such as harmful microorganisms (Kamra *et al*., 2012) [6]. There are about 200,000 PSM structures that have been recognized (Patra & Saxena, 2010) [7]. The majority of PSMs are divided into three major categories: saponins, tannins and essential oils (EOs).

EOs are blend of secondary metabolites produced from the plant volatile fraction by solvent extraction and steam distillation and have antimicrobial properties (Gershenzon and Croteau, 1991; Rira *et al*., 2015) [5, 10]. Among the EOs, the ajwain (*Trachyspermum ammi*) oil is one of the very popular feed additives for livestock. Ajwain seeds contain 2.0 to 4.4% oil that contains thymol (35-60%), a monoterpene with antimicrobial potency due to the presence of a hydroxyl group in the phenolic structure (Ultee *et al*., 2002). At low doses, EOs suppresses methane production, but at greater concentrations, they may have a detrimental effect on fibre utilization (Pawar *et al*., 2014). Bahera (*Terminalia bellirica*) is an ayurvedic medicinal plant and is a rich source of tannin. Tannins are water-soluble phenolic compounds (M. wt. 500-3,000 D) and contain many hydroxyl and other functional groups (1-2 per 100 D). They can establish cross-links with proteins and increase protein digestibility (Bate-Smith, 1972) [17].

Keeping in mind, the credibility of ajwain oil and bahera as potent feed additives, the present study was planned to study the effect of ajwain oil and bahera combination nutrient utilization in utilization in dairy cattle.

Material and Methods

The experiment was carried out in Livestock Production Management Section, ICAR-IVRI, Izatnagar. Eighteen, lactating cross bred Vrindavani cattle (30 to 60 days postpartum) were selected (based on parity and milk yield) and divided into 3 groups with each group containing 6 animals based on RBD (Randomised Block Design). Control group was fed with basal diet (concentrate according to milk yield and green fodder). In T_1 group, basal diet was supplemented with a combination of ajwain oil and bahera. In T_2 group basal diet was supplemented with a combination of ajwain oil and bahera at double the dose of T_1.

Feeds and feeding

Concentrate mixture requirement of animals was calculated according to their milk yield. The calculated concentrate mixture amount was divided into three parts and offered to animals during milking hours. Additives were mixed in the concentrate mixture. After the concentrate was completely consumed, the green fodder and wheat straw were offered *ad libitum*. The experimental animals were offered clean and fresh drinking water.

Digestion trial

Acid-insoluble ash was considered a natural indicator for measurement of nutrient digestibility (Shrivastava and Talapatra, 1962) [11]. Weighed quantities of feeds were offered to animals twice daily at 9.00 AM and 3.00 PM for 5 days. During the collection periods, representative samples of feed offered and faeces were collected from each group. The respective samples of faeces were collected in sample bags at 12 PM, 2 PM, 5PM, 7PM and 9 PM to offset the possible diurnal variation. All the collected samples were kept for dry matter at 70 °C for 24 hours. Proximate analysis and fibre analysis of feed sample were done as per the methods of AOAC (1995) [1] and Van Soest *et al*. (1991) [14] respectively.

Statistical analysis

The results obtained were statistically analysed as per Snedecor and Cochran, (1889) [12] using IBM, SPSS (26.0). Data were analysed by one-way ANOVA. Means were compared implying Tukey's test and the $p<0.05$ was referred as significant difference.

Results

Effect of plant secondary metabolites supplementation on nutrient intake and digestibility in cross bred dairy cattle

Data pertaining to dry matter intake (DMI) during the digestion trial and digestibility of different nutrients is depicted in Table 1. There was no significant ($p<0.05$) effect of additive supplementation on DMI (kg/day) during the digestion trial with mean value 13.99, 13.91 and 13.87 in control, T_1 and T_2 groups respectively. The mean value of dry matter digestibility (%) of control, T_1 and T_2 was 64.79, 65.64 and 66.52 respectively. Digestibility of nutrients CP, OM, ADF and EE were comparable among groups whereas neutral detergent fibre (NDF) digestibility was significantly higher ($p<0.05$) in additive-supplemented groups than control.

Table 1: Effect of plant secondary metabolites on DMI and nutrient digestibility

Attributes	Control	T_1	T_2	SEM	P-value
Nutrient Intake (kg/day)					
Dry matter	13.99	13.91	13.87	0.40	0.992
Organic matter	12.67	12.58	12.57	0.37	0.994
Crude protein	1.89	1.89	1.90	0.07	0.999
Neutral detergent fibre	6.65	6.52	6.39	0.11	0.666
Acid detergent fibre	3.31	3.25	3.18	0.05	0.553
Ether extract	0.43	0.42	0.43	0.02	0.896
Nutrient digested (kg/day)					
Dry matter	9.08	9.14	9.30	0.36	0.970
Organic matter	8.55	8.56	8.73	0.33	0.973
Crude protein	1.31	1.35	1.38	0.07	0.914
Neutral detergent fibre	3.62	3.63	3.66	0.08	0.985
Acid detergent fibre	1.611	1.53	1.56	0.03	0.511
Ether extract	0.34	0.33	0.33	0.01	0.967
Nutrient digestibility%					
Dry matter	64.79	65.64	66.52	0.63	0.555
Organic matter	67.40	67.95	68.96	0.58	0.562
Crude protein	67.04	71.11	72.02	0.83	0.345
Neutral detergent fibre	54.48^{a}	55.71^{ab}	57.09^{b}	0.36	0.005
Acid detergent fibre	48.72	47.24	48.89	0.25	0.067
Ether extract	75.72	74.10	74.99	0.58	0.549

Control No: Additive supplementation, T_1 - diet supplemented with combination of ajwain oil and bahera, T_2 -basal diet was supplemented with a combination of ajwain oil and bahera at double the dose of T_1

Effect of PSMs supplementation on plane of nutrition

Body weight, metabolic body weight (kg) and feed intake of all experimental groups during digestion trial are presented in Table 2. Body weight and metabolic body weight was similar in all experimental group. Roughage intake and concentrate intake for control, T_1 and T_2 was 6.17, 5.91, 5.73 and 7.82, 7.73, 7.75 respectively.

The digestible crude protein (DCP) intake (kg/day) was comparable ($p>0.05$) in all experimental groups with mean values of 1.28, 1.29 and 1.32 for control, T_1 and T_2 group respectively. TDN intake in terms of kg/day and gm/kg$W^{0.75}$ was also comparable ($p>0.05$) in all treatment groups.

Table 2: Feed intake and plane of nutrition in cross-bred dairy cattle

Attributes	Control	T_1	T_2	SEM	p-value
Body weight(kg)					
Body weight	388.3	428.3	431.7	11.86	0.265
$W^{0.75}$	87.33	94.09	94.62	1.97	0.256
Feed intake (kg/day)					
Roughage intake	6.17	5.91	5.64	0.05	0.067
Concentrate intake	7.82	7.88	8.01	0.39	0.996
g/kg$W^{0.75}$	161.6	144.1	146.9	4.82	0.412
% body weight	3.66	3.26	3.23	0.12	0.289
DCP intake					
kg/day	1.31	1.34	1.38	0.07	0.914
g/kg$W^{0.75}$	15.07	14.31	14.62	0.71	0.918
TDN Intake					
kg/day	9.01	9.01	9.19	0.35	0.976
g/kg$W^{0.75}$	103.96	96.01	97.21	3.77	0.678
Nutrient density%					
DCP	9.30	9.61	9.79	0.20	0.608
TDN	64.28	64.74	65.71	0.56	0.591

Control no: additive supplementation, T_1 - diet supplemented with combination of ajwain oil and bahera, T_2 -basal diet was supplemented with a combination of ajwain oil and bahera at double the dose of T_1

Discussion

The dry matter intake (DMI) (kg/day), nutrient intake (kg/day) and digestibility (%) of OM, CP, ADF and EE were similar (p>0.05) for all experimental groups whereas NDF digestibility was significantly improved in T_2 group. In the same manner Benchaar *et al*. (2003) [3] found no significant difference in DMI by supplementation of combination of essential oil (Crina Ruminants@ 750 mg/day) in lactating dairy cows. Similarly, Yatoo *et al*. (2018) [16] reported that the addition of the essential oil blend (equal proportion of ajwain oil, garlic oil and cinnamon leaf oil) at 0.5 and 0.30 ml/kg of DMI did not appear to have a noticeable impact on DMI and nutrient digestibility in growing buffaloes. Wadhwa and Bakshi, (2019) [15] also reported that the supplementation of ajwain oil at the dose rate of 0.05% of DMI did not affect the daily DMI and nutrient digestibility in buffalo calves. Contrary to the present findings, Patra *et al*. (2011) [8] observed increased nutrient digestibility by supplementation of harad and garlic to sheep when it was added at the dose rate of 1% DMI. In the present study improvement in NDF digestibility may be attributed to the ability of plant secondary metabolites (PSMs) to augment the activity of fiber-digesting enzymes.

Plane of nutrition

During metabolic trial the average body weights of all four experimental groups showed no significant variations. The intake of CP and TDN (kg/kg$W^{0.75}$) were also found comparable (p>0.05) in all three experimental groups indicating the animals of all three groups during the digestion trial were in similar plane of nutrition. Similarly, Yatoo *et al*. (2018) [16] reported that blend of essential oil at the dose rate of 0.15 and 0.30 ml/kg of DMI did not affect body weight change and plane of nutrition. Patra *et al*. (2011) [8] observed no impact on plane of nutrition by supplementation of harad and bulb of garlic individually or in combination at 1% of DMI in sheep.

Conclusion

Results from the experiment indicate that the dietary supplementation of plant secondary metabolites did not affect nutrient intake nutrient digestibility in cross-bred dairy cattle except NDF digestibility which was improved.

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