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Role of *Aloe barbadensis* supplementation on haematological parameters and rumen development of crossbred calves

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Abstract

The present experiment was conducted to study the effect of *Aloe barbadensis* supplementation through mixing in milk on hematological and certain rumen fluid parameters of crossbred calves. For this purpose, a 56 day's feeding trial was conducted on growing crossbred calves. Aloe vera (*Aloe barbadensis*) was supplemented at graded levels as powder of leaf of Aloe vera (2 g/Kg body weight and 4 g/ Kg body weight) through milk to treatment groups T1, and T2, respectively with 3 replicates of 12 crossbred calves, each for a period of 56 days. The overall mean haemoglobin, packed cell volume percentage, total leucocyte count, neutrophil and lymphocyte number differed significantly (P<0.05) between aloe vera supplemented groups and control. Total volatile fatty acids (mEq/dl) in Aloe vera supplemented group were significantly higher at 42th day and 56th day of experimental trial compared to control group. The T₂ supplemented group had significantly lower ruminal bacterial populations than control at 42nd and 56th of experiment. Rumenal bacterial population was highest and lowest in T1 and T2 groups respectively. The study revealed that *Aloe barbadensis* supplementation of aloe vera in milk would be beneficial in increasing blood profile (Hb, PCV, TEC and TLC), total volatile fatty acids concentration helps in early development of rumen.

Keywords: crossbred calves, haematology, rumen, volatile fatty acid

1. Introduction

Better care and management of calves are helpful as replacement herd in both dairy and beef production system^[1]. A calf requires nutritionally balanced diet therefore, feed is an important and critical input for the calves in sustainable dairy industry, as it accounts for 60 to 70 percent of production cost. Earlier many feed additives like antibiotics and hormones like diethyl stilbesterol were used for calves as growth promoters. But due to worldwide denouncement over their use as growth promoters lead to increased demand of other feed supplements. Various herbal plants have been used to improve growth rate, feed efficiency and blood profile and to reduce the production cost. The main objective of adding feed supplement is to boost calves performance by increasing their growth rate, better feed conversion efficiency, greater livability and lowered mortality in calves. Among all the medicinal plant, aloe vera is one of the most important medicinal plants which have several medicinal and therapeutic properties, therefore it is known as miracle plant. Aloe barbadensis, also called Aloe vera, and Aloe arborescence are the most commonly grown species of Aloe vera. Polysaccharides, vitamins, anthraquinones, enzymes and low molecular weight compounds are the major components ^[16] of Aloe vera. It has multiple health benefits viz, immunomodulatory, antiinflammatory, antiviral, antifungal, antitumor, wound healing, antidiabetic and antioxidant effects ^[17]. Little work has been done on aloe vera supplementation in calves. So keeping in view of the above, present experimental study was designed with an aim to evaluate efficacy of two different levels of Aloe vera supplementation on blood and rumen fluid profile in dairy crossbred calves.

2. Materials and Methods

Experimental animals: An experiment was conducted on twelve newborn female crossbred calves at Instructional Dairy Farm, Nagla, G. B. Pant University of Agriculture and Technology, Pantnagar, Uttarakhand to assess the effect of Aloe vera supplementation on feed

In take efficiency, growth rate and internal parasitic load. The growth trial was conducted for 9 weeks including first week as acclimatization feeding period. A total of twelve crossbred (Holstein Friesian × Sahiwal) female calves (5-day-old) weaned after birth and were divided into three groups of four animals in each group on the basis of their body weight as TO (Control), T1 (Aloe vera supplementation @ 2 g per kg body weight), T2 (Aloe vera supplementation @ 4 g per kg body weight. All the calves were kept in well ventilated shed and fed individually in calf pens. They were kept in the open paddock during the first half of the day for getting sunshine and exercise. Experimental shed and open paddock were disinfected with 1 per cent phenyl solution before housing the calves. Roughages and calf starter were fed to all the groups of crossbred calves both in the morning and evening hours from 15th day onwards. Ad libitum water was provided twice a daily at 8 AM and 3 PM to the animals throughout the experimental period. The milk was first boiled and then cooled to body temperature before feeding. The quantity of whole milk to be fed to each calf was divided into two equal halves and was fed in morning and evening.

2.1 Description of Aloe vera (procurement, processing and feeding)

Aloe vera leaves were procured from Medicinal Research Development Center (MRDC) situated at outskirts of G. B. Pant University of Agriculture and Technology, Pantnagar. Procured Aloe vera leaves were subjected to thorough washing and then dried in the shed before processing. These leaves were minced in the electric operated mixer to prepare the crude extract so that loss of some active ingredients like enzymes, hormones and vitamins could be avoided. Procurement and processing of Aloe vera were done daily. Now this processed aloe vera was fed to the treatment groups of animals twice a day at 4.30 AM and 4.30 PM throughout the experimental period. Since data of weight gain were taken weekly, therefore mean of the body weight of treatment groups were considered for calculation of amount Aloe vera. Once amount of Aloe vera for a group was calculated then it was continued to fed till next due date of data collection.

2.2 Sampling

The samples of blood and rumen fluid were collected during winter season (February to March).

2.3 Collection of blood samples

Blood samples were collected from all experimental animals on 0 day (pre trialsamples), 14th day and 28th day (mid trial), 42th day and 56th day (end trial) of the feeding trial. Blood was collected from the calves for hematology parameters estimation. For hematological examination, 1 to2 ml of blood sample were drawn from jugular vein and EDTA as anticoagulant was added @ 1 mg/ml for hemoglobin (Hb), packed cell volume (PCV), total leukocyte count (TLC) and differential leukocyte count (DLC) parameters and vials were stirred slowly on palm for proper mixing of EDTA with blood. Immediately after collection, the test tubes were kept in thermacool boxes in standing position. Immediately after collection of blood, blood sample collected in vials for hematological parameters were carried to the laboratory for analysis. Haemoglobin was estimated by Sahil's method^[2]. Packed cell volume was done by Wintrobe^[3] method. Total erythrocyte and total leukocyte count were done by hemocytometer and Schalm et al. [4] method respectively.

Rumen fluid: 10-15 ml rumen liquor was sampled from the crossbred calves by rumenocentesis with the help of (18-20 gauge length 6 inch) needle for ruminal fluid analysis. Samples were centrifuged at 2000 rpm for 5 min and clear supernatants were obtained. Two drops of saturated Hgcl₂ were added to the supernatant of rumen liquor as preservative. The strained rumen liquor (SRL) was centrifuged for one minute at 1000 rpm to remove heavier and suspended food particles.

2.4 Determination of total volatile fatty acids (TVFA)

Total volatile fatty acid was determined by the method of Annison^[5].

2.5 Total bacterial count

Total bacterial counts were determined by the method as described by Gall *et al.* ^[6].

2.6 Statistical analysis

The experimental data obtained during the study were analyzed statistically using completely randomized design with the simple analysis of variance technique. Weekly and fortnightly mean with standard error where estimated for respective treatments separately. The difference among the treatment means for each character was further tested ^[7].

3. Results and Discussion

3.1 Effect of *Aloe vera* supplementation on haematological parameters

The effect of Aloe vera supplementation on hemoglobin concentration, packed cell volume percentage, total leucocyte count, total erythrocyte count, neutrophil and lymphocyte number are presented in table 1. The mean haemoglobin, packed cell volume percentage, Total leucocyte count, neutrophil and lymphocyte number across the period differed significantly (P < 0.05). The hemoglobin value in Aloe vera supplemented group compared to control group showed significant difference (P < 0.05) during 42th days and 56th day post feeding of Aloe vera. The packed cell volume percentage value in Aloe vera supplemented group compared to control group showed significant difference (P < 0.05) at 28th day and 42th day post feeding of Aloe vera. At 56th day of observation packed cell volume percentage value in Aloe vera supplemented group compared to control group showed significant difference (P < 0.01) also. The total erythrocyte count (10⁶/µl) and total leucocyte count (10³/µl) at 42th days and 56th day of observation total erythrocyte count $(10^{6}/\mu l)$ value in Aloe vera supplemented group significant difference (P < 0.05) also. The neutrophil and lymphocyte number value in Aloe vera supplemented group compared to control group showed significant difference (P < 0.05) from 14 days post feeding of Aloe vera. The increase in haemoglobin content might be due to antioxidant property of Aloe vera as its components compete with hemoglobin in red blood cells for oxygen, resulting in hypoxia, which then stimulates the Hemoglobin synthesis and red blood cells production. The increase in hematological parameters may also be attributed to the fact that Aloe polysaccharide contains acemannan, which is a β -linked acetylated mannose of approximately 1000 kilo Dalton molecular weight. The biological activity of mannose is activation of macrophages and stimulation of T-cells. Acemannan stimulates the antigenic response of lymphocytes. Aloe vera is also a bone marrow stimulant. It also stimulates the formation of all types of cells from both the spleen and the

bone marrow, resulting in an increase in the hematological parameters. The results of present study were in conformity with the results of Bombik et al. [8] who reported significantly higher red blood cell count, haemoglobin concentration, and corpuscular volume in calves supplemented with different herbs. However in their study significantly lower white blood cell counts were found. Singh et al. [9] reported significantly higher values of Hb concentration, PCV percentage and total leukocyte count in broiler chicken supplemeted with Aloe vera. Dar *et al.* ^[10] reported that there were better haematological parameters (haemoglobin, PCV, TEC, TLC, lymphocyte percentage) in crossbred calves supplemented with probiotic, prebiotic and symbiotic. However no difference was observed in neutrophil percentage. Also there was no significant difference in the haematocrit values between the supplemented and control group. However contrary to our results, Bhati [11] reported that there was no effect of feeding aloe vera on haemoglobin and PCV content of crossbred calves.

3.2 Effect of Aloevera supplementation on total volatile fatty acids

Effect of Aloe vera supplementation on total vollatile fatty acids (mEq/dl) are presented in table2. The mean total volatile fatty acids (mEq/dl) level in rumen fluid were 80.1, 85.65, 84.78 in group T0, T1 and T2 respectively and the value differ significantly (P < 0.05), also numerically higher values were observed in group supplemented with Aloe vera supplementation @ 4 g per kg body weight (i.e. T2 group), followed by Aloe vera supplementation @ 2 g per kg body weight group (i.e. T1 group). The peak total volatile fatty acids (mEq/dl) in rumen fluid appeared at 56th day in the groups and before it the value increases with age of calves. At 42th day and 56th day of observation total volatile fatty acids (mEq/dl) in Aloe vera supplemented group were significantly higher compared to control group. The results of present study are consistent with the study of Calabro et al. [12] who reported Aloe Arborescens significant increased in-vitro TVFA concentration. However, contrary to our study, Wanapat et al. [13] reported that there was no effect of plant herb combination supplementation on TVFA concentrations in beef cattle.

3.3 Effect of Aloevera supplementation on bacterial count

Effect of Aloe vera supplementation on bacterial count (10⁸) are presented in table 2 The mean of bacterial count (10^8) level in rumen fluid were 1.78, 1.86, 1.62 in T0, T1 and T2 groups respectively and the value differ significantly (P < 0.05), also numerically lower values were observed in group supplemented with Aloe vera supplementation @ 4 g per kg body weight (i.e.T2 group), followed by Aloe vera supplementation @ 2 g per kg body weight group (i.e. T1 group). Bacterial count (10⁸) ranged from 1.78 to 2.19 at 56th day of observation among the groups and the values were statistically similar. The peak bacterial count (10^8) in rumen fluid appeared at 56th days in the groups and before it the value increased with age of calves. The T₂ supplemented group had significantly lower ruminal bacterial populations than control at 42nd and 56th of experiment. Wanapat et al. ^[13] also reported significantly lower bacterial titre in rumen in animals supplemented with herbs. Contrary to our using peppermint oil or garlic powder diets, ^[14] and Kongmun *et al.* ^[15] reported that the numbers of cellulolytic bacteria increased.

Table 1: Effect of feeding different dietary levels of Aloe vera on	
haematological parameters of crossbred calves.	

Period (Days)						M	CT.				
Groups	0	14	28	42	56	Mean	SEm				
Haemoglobin (g/dl)											
T_0	9.6	9.8	10.02	10.06 ^a	10.38 ^a	9.972ª	0.537				
T_1	10.05	11.05	11.97	12.7 ^b	13.37 ^b	11.82 ^b	0.754				
T_2	9.8	10.75	11.25	13.2 ^b	14.31 ^b	11.862 ^b	0.119				
PCV											
T_0	28.75	29.08	29.4 ^a	29.88ª	29.95 ^a	14.963 ^a	2.405				
T_1	28.75	30.7	36.61 ^b	39.85 ^b	41.9 ^b	20.628 ^b	2.325				
T_2	28	31.3	35.55 ^b	40.75 ^b	41.61 ^b	20.666 ^b	0.21				
			TEC(1	.0 ⁶ /µl)							
T_0	6.74	6.90	7.14	7.15 ^a	7.18 ^a	7.022	0.279				
T_1	7.0	7.45	7.59	8.12 ^b	8.35 ^b	7.702	0.22				
T_2	6.61	7.57	7.71	8.18 ^b	8.36 ^b	7.686	0.079				
			TLC(1	$0^{3}/\mu$ l)							
T_0	8.6	8.6	11.08	10.63 ^a	12.36 ^a	10.25 ^a	1.223				
T_1	8.57	9.48	11.68	12.73 ^b	13.39 ^b	11.17 ^b	0.848				
T_2	8.67	9.49	13.12	14.82 ^b	15.1 ^b	12.24 ^b	0.669				
			Neutro	ophils							
T_0	25.75	13.25 ^a	13.5 ^a	12.5 ^a	13.5 ^a	15.7 ^a	1.68				
T_1	29.5	25.5°	27.5°	28.5°	29.35 ^b	28.07 ^c	0.671				
T_2	27	18.25 ^b	20 ^b	23 ^b	27.5 ^b	23.15 ^b	2.3				
Lymphocytes											
T_0	63.0	69.25 ^a	72 ^a	70.75 ^a	70.5 ^a	69.1ª	2.206				
T_1	65.5	78.75 ^b	77.5 ^b	75.25 ^a	71ª	73.6 ^a	1.448				
T_2	67	86.75 ^c	82 ^b	84.5 ^b	83.5 ^b	80.75 ^b	3.216				

Means bearings different superscripts in a column differ significantly (p < 0.05)

 Table 2: Effect of feeding different dietary levels of Aloe vera ontotal vollatile fatty acid concentration and Bacterial count of crossbred calves

Groups		Perio	Mean	SEm±						
	14	28	42	56						
Total vollatile fatty acids (meq/l)										
T0	63.18	69.47	95.3 ^b	92.45 ^b	80.1 ^b	7.97				
T1	64.2	71.58	99.85 ^a	106.97ª	85.65 ^a	8.577				
T2	65.45	71.5	98.4ª	103.75 ^a	84.78 ^a	6.561				
Bacterial Count (x10 ¹⁰ /ml)										
T0	1.55	1.68 ^a	1.86 ^a	2.02 ^a	1.78 ^a	0.069				
T1	1.59	1.73 ^a	1.94 ^a	2.19 ^a	1.86 ^a	0.116				
T2	1.45	1.57 ^b	1.69 ^b	1.78 ^b	1.62 ^b	0.095				

Means bearings different superscripts in a column differ significantly (p < 0.05)

4. Conclusion

The study revealed that *Aloe barbadensis* supplementation of aloe vera in milk would be beneficial in increasing blood profile (Hb, PCV, TEC and TLC), total volatile fatty acids concentration and bacterial count in cross bred calves. Thus *Aloe barbadensis* supplementation helps in early development of rumen.

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