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Methane Mitigation Potential Assessment of Herbal Lean Meat Product in Deccani Sheep

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ABSTRACT

Methane has global warming potential and also accounts for a significant energy loss to the ruminant animal. The trial was conducted to study the efficacy of dietary supplement herbal formulation AV/LMP/10 (herbal lean meat product) (*M/S Ayurvet Limited, India*) on methane mitigation potential, digestibility and nutrient utilization in Deccani sheep. 24 growing healthy Deccani sheep (5-6 months) were randomly divided into 2 groups. Control Group T_0 was not given any treatment. Group T_1 supplemented with AV/LMP/10@1kg/tonne of feed for 3 months. Parameters viz. methane emission estimation, body weight, plasma cortisol level, liver marker enzyme estimation, feed analysis, nitrogen balance, and carcass quality traits were studied. The AV/LMP/10 supplemented group T_1 emitted significantly ($P<0.05$) less methane as compared to control group T_0. Statistically ($P<0.05$) AV/LMP/10 supplemented group had significantly superior average daily gain (g) than that of control. DMI, Dry matter digestibility and FCR of AV/LMP/10 supplemented group were significantly ($P<0.05$) improved. The maximum nitrogen balance was attained in AV/LMP/10 supplemented group T_1. Significantly ($P<0.05$) lower level of cortisol in AV/LMP/10 supplemented animals confirmed the increased resilience of animal towards stress. Animals belonging to AV/LMP/10 supplemented group T_1 had significantly higher dressing % as compared to control group T_0. The liver marker enzyme levels in both groups were under normal physiological level. Herbal lean meat product AV/LMP/10 was found to be safe for usage and has efficient methane mitigating potential along with added benefit of improvement in digestion, nutrient utility, performance traits in sheep.

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Introduction

Livestock contribute in emission of greenhouse gases such as carbon dioxide, methane and nitrous oxide both directly and indirectly[1]. The relative contribution to CH_4 emissions by ruminants was 93.4%, as compared with 6.6% by non-ruminants[2]. With rapid transformation from traditional agriculture to industrial farming, modern livestock husbandry is rapidly expanding. 40% globally and 30% in the developing countries, the livestock contributes to the agricultural gross domestic product[3]. Sheep have significant role in economy. According to the 1972 census, the country had 40 m sheep, which contributed approximately $175 m (Rs 1400 m) per year to the national economy, based on a rough estimate of production of 34.3 m kg of wool, 101 m kg of mutton, and 14.6 m skins[4]. Because of economical advantage and increasing concerns for health lead the efforts to develop new foods with positive health benefits[5]. Meat is still the most valuable source of high-value animal protein worldwide comprises mostly monounsaturated and SFAs[6]. Meat supplies about one third of the dietary cholesterol in many western diets[7]. In most industrialized countries, a high meat intake contributes to a higher than recommended total and saturated fat and cholesterol intake but on the other hand meat may replace sources of other important nutrients in the diet. Therefore, the nutrition expert advice to prefer lean meats and low-fat meat products only[8]. Small ruminant muscle had higher PUFA: SFA ratio than those reported for beef, which may be important in human nutrition[9].

Because of increased animal protein demand, the use of modern rearing techniques in husbandry is also increased. Because of this the population of livestock is increasing along with the threat of increased emission GHGs from the livestock. AV/LMP/10 is blend of scientifically validated herbs for production of low fat meat with better organoleptic properties. The current study was conducted to evaluate the methane mitigation potential of AV/LMP/10 along with its effect on meat quality.

Materials and Methods

Present study was undertaken on Deccani sheep at Livestock Farm, Hayathnagar Research Farm (HRF), Central Research Institute for Dryland Agriculture (CRIDA), Hyderabad, India and was situated at 17°27'N latitude and 78°35'E longitude and about 515 m above sea level. The climate is semi-arid with hot summers and mild winters. The mean maximum air temperature during summer (March, April and May) ranges from 35.6 to 38.6°C, whereas, in winter (December, January and February) ranges from 13.5 to 16.8°C.

Experiment design

12 growing healthy Deccani sheep of nearly same age (5-6 months) and body weight (Kg) 16.35 to 16.48 were selected for the study. These animals were randomly divided into two groups, so that each group was having similar and uniform age and body weight. The animals were kept in two pens to acclimatize to the conditions for a period of 7 days prior to commencement of animal experimentation. Group T_0 (n=6) was not given any treatment and served as control. Group T_1 (n=6) was supplemented with AV/LMP/10@1kg/tonne of feed for 3 months along with concentrate mixture. Concentrate mixture was made from maize, groundnut cake, wheat bran, rice polish, mineral mixture and salt and offered to each animal @ 250 gms daily. Green fodder of Hybrid Napier (Co-4 variety) was offered fresh in the pen after thorough chaff cutting @ 4 Kg per day. Hygiene conditions were maintained each day, the floor of the

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experimental shed was cleaned with potassium permanganate, a sanitizer. During the trial parameters viz. methane production estimation, body weight gain/week, analysis of food/faeces samples, plasma cortisol level, blood enzyme estimation, phenolic compound estimation, and carcass quality traits were studied.

Statistical analysis

Statistical analysis of the scientific data collected during the experiment was done with method described by Snedecor and Cochran[10]. Results obtained from the present investigation were summarized on basis of Mean ± SE at 5% level of significance.

Results and Discussion

Methane emission estimation

Enteric emissions from the animals were measured using closed respiratory chamber method (Fig. 1). Air samples from the chamber were collected from various heights at regular interval of 60 min in 24h duration. After sampling, gas samples were analyzed on same day for methane concentration using a gas chromatograph (450-GC, BRUKER Daltonics, Bremen, Germany) with three detectors Thermal Conductivity Detector (TCD), Electron Capture Detector (ECD) and Flame Ionization Detector (FID) with a 1041 PWOC Packed/Wide bore On-Column[11]. In the present investigation, AV/LMP/10 supplemented T_1 group was found to be significantly ($p<0.05$) efficient in reducing methane emission (Methane weight emitted gms/kg of DDMI is 41.580[b]) in comparison of control T_0 group (Methane weight emitted gms/kg of DDMI is 47.322[a]) (Table 1).

Methane (CH4) promotes stratospheric ozone depletion[12]. Methane is the second major contributor to global warming with a 100-year global warming potential (GWP), 23 times that of CO_2[13]. Methane accounts for a significant energy loss to the ruminant animal, amounting to about 8% of gross energy at maintenance level of intake and falling to about 6% as the level of intake rises[14]. Low emission of CH4 in the rumen with AV/LMP/10 supplementation has implications not only for global environmental protection but also for efficient animal production.

Plant secondary metabolites such as saponins and tannins have a role in reducing CH_4 emissions[15]. Saponins have been shown to possess strong defaunating properties both in vitro[16] and in vivo, which could reduce CH_4 emissions. Recently, a number of studies have reported that feeding tannin-containing forages to ruminants may reduce methane emissions[17-19]. In the present experiment also, significantly ($p<0.05$) maximum pure tannin and CT content was found AV/LMP/10 supplemented T_1 group (Table 2) which explains the maximum reduction in methane emission by this group of animals.

Plan of nutrition

When energy and protein intake as per dry matter intake (DMI) was extrapolated, it was observed that supplemented group animals consumed significantly ($p<0.05$) more TDN, DCP, DE and ME which along with added advantage of superior digestibility resulted into attainment of better body weight as compared to control (Table 3). Group T_1 supplemented with AV/LMP/10 consumed significantly ($p<0.05$) more TDN, DCP, DE and ME. This might be due to individual herbal ingredients of AV/LMP/10 viz. *Commiphora mukul, Allium sativum & Trigonella foenum graecum* which were scientifically validated to stimulate the digestive function, better feed assimilation and metabolism[20,21].

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DMI and FCR

Dry matter intake (g/d) (Table 4) in treatment group T_1 (726.83 ± 15.18^b) was significantly ($p<0.05$) higher than control group T_0 (698.71 ± 12.12^a). This may be attributed to key herbal ingredients of AV/LMP/10 that might have improved palatability of the feed as well as nutrient utilization.

Feed conversion ratio of AV/LMP/10 supplemented T_1 group was significantly ($p<0.05$) superior (14.63 ± 1.23^b) than control T_0 group (16.64 ± 1.21^a), which suggests supplementation of AV/LMP/10 to be efficient in converting feed to meat. The similar findings were observed in broiler by Sahoo *et al* on AV/ LMP/10 supplementation[22].

Digestibility coefficients

One week of digestibility trial suggested that DM, OM, CF, CP, EE, NDF and ADF digestibility was significantly ($p<0.05$) better in AV/LMP/10 supplemented group T_1 as compared to control group T_0 (Table 5). This improvement in nutrient digestibility leads to better availability of nutrients which explains the superior average daily gain and achieving greater body weight of AV/LMP/10 supplemented group as compared to control.

Average daily weight gain

The results of average daily weight gain (gms/d) are presented in Table 6. Group T_1 animals supplemented with AV/LMP/10 revealed significant ($p<0.05$) increase in weight gain over control group at 30th day of the treatment. After three months of treatment the average daily weight gain (gms/d) in AV/LMP/10 supplemented group T_1 (49.64 ± 7.74^b) was significantly ($p<0.05$) higher as compared to control group T_0 (42.07 ± 5.69^a). The results are in accordance with the findings of Mane *et al*[23] who observed significant increase in average daily weight gain in broilers after AV/LMP/10 supplementation for 0-6 weeks. This may be attributed to the efficacy of constituent herbs of AV/LMP/10 namely *Commiphora mukul*, *Trigonella foenum graecum*, *Allium sativum* & many more which are scientifically well proven for improving growth, productivity & hepatoprotective action[24].

Nitrogen balance

When nitrogen balances through the metabolic trial results were estimated, It was observed that significantly ($p<0.05$) better nitrogen balance was attained by AV/LMP/10 supplemented group T_1 (Nitrogen balance, 4.94 ± 0.21^b g/day) in comparison of control group T_0 (Nitrogen balance, 3.33 ± 0.12^a g/day) (Table 7). More is the retention of nitrogen lower is its release into the environment, better is protein retention.

Blood cortisol level

Cortisol is released in response to stress. Anxiety-related behavior of cattle can be correlated with cortisol levels[25]. Blood cortisol level was estimated in the both groups at different time intervals. Initially cortisol level was significantly less ($p<0.05$) in control group T_0 animals than supplemented animals (Table 8). But the trend was totally reversed after supplementation where AV/LMP/10 supplemented group T_1 animals had less cortisol value (25.73 ± 1.56^b) than control (33.12 ± 2.02^a), which suggests that herbal formulation used in the experiment has increased the tolerance capacity of animals against heat stress.

Liver marker enzymes levels

The over all alkaline phosphatase level (U/L) in control group T_0 (176 U/L) and in AV/LMP/10 supplemented group T_1 (202 U/L) was found to be in normal range though numerically higher in supplemented group. ALP has a role in detoxification of endotoxin. So high level of ALP in supplemented group within physiological limit, made animals more endurable against toxins. The serum SGOT level was found to be 91 U/L in AV/LMP/10 supplemented group T_1, the values were significantly less than 124 U/L i.e of control group T_0. The plasma SGPT level was significantly less in AV/LMP/10 supplemented group T_1 i.e. 17.6 U/L in comparison to untreated control group T_0, 27 U/L. The low level of SGOT and SGPT within physiological limits is a favourable adaptive status against stress.

Carcass quality traits

At the end of the 90-day feeding trial, all animals were slaughtered for carcass characteristics evaluation. When animals were slaughtered for carcass characteristics, it was found that animals belonging to AV/LMP/10 supplemented T_1 group (49.3 ± 3.01^b) had ($p<0.05$) significantly higher dressing % (Table 9) as compared to control group T_0 (44.1 ± 1.02^a), which suggests superior utilization of feed resources for production of edible parts in supplemented groups with AV/LMP/10. The herbs of AV/LMP/10 viz. *Trigonella foenum graecum* and *Allium sativum* were scientifically proven to be effective in improving carcass quality[26,27].

Conclusion

AV/LMP/10 is a scientifically developed non hormonal herbal blend useful in producing low fat meat with better organoleptic properties. From the results of the present investigation it can be concluded that the herbal formulation AV/LMP/10 feeding not only improved the digestibility, weight gain, health, carcass traits of the sheep but it can be concluded that AV/LMP/10 is a environmental friendly product with significant methane mitigation potential.

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References

1. Johnson KA, Johnson DE. Methane Emissions from Cattle. *J. him Sci*. 1995; 73: 2483-2492.
2. Lianga L, Lal R, Duc Z, Wua W, Menga F. Estimation of nitrous oxide and methane emission from livestock of urban agriculture in Beijing. *Agri Ecosyst Environ*. 2013; 170: 28– 35.
3. World Bank. Minding the stock: Bringing public policy to bear on livestock sector development. Report No. 44010-GLB. The World Bank, Washington D.C., USA; 2009.
4. NCA. Report of the National Commission on Agriculture, Part VII: Animal Husbandry. Govt. of India, Ministry of Agriculture and Irrigation, New Delhi; 1976.
5. Mallika EN, Prabhakar K, Reddy PM. Low fat meat products-an overview. *Vet world*. 2009; 2(9): 364-366.
6. Halwai A. Replacement of Saturated Animal Fats in Meat Products: A Review. *J Food Sci. & Technol*. 2012; 7: 9-13.
7. Bender A. Meat and meat products in human nutrition in developing countries. FAO. Food and Nutrition paper 53. Food and agriculture organization of the United Nations. Rome; 1992.
8. Valsta LM, Tapanainen H, Männistö S. Meat fats in nutrition. *Meat Sci*. 2005; 7(3): 525–530.
9. Young WP, Washington AC. Fatty acid composition of goat organ and muscle meat of alpine and Nubian breeds. *J food sci*. 1993; 58(2): 245-248.

10. Snedecor GW, Cochran WG. Statistical methods. 6h ed. Amos Lowa, U.S.A: The Lowa State University Press; 1991. p. 1-503.
11. Sitaula BK, Luo J, Bakken LR. Rapid analysis of climate gases by wide bore capillary gas chromatography. *J Environ Quality*. 1992; 21: 493-496.
12. Blake DR, Rowland FS. Continuing worldwide increase in tropospheric methane 1978 to 1987. *Sci*. 1988; 239: 1129-1131.
13. IPCC. Climate Change 2001. The Scientific Basis. In: Contribution of working group I to the Third Assessment Report of the Intergovernmental Panel on Climate Change, New York: Cambridge University Press; 2001. p. 1-873.
14. Bhatta R, Enishi O, Kurihara M. Measurement of Methane Production from Ruminants. *Asian Aust J Anim Sci*. 2007; 20: 1305-1318.
15. Wallace RJ. Antimicrobial properties of plant secondary metabolites. *Proc Nutr Soc*. 2004; 63: 621-629.
16. Wallace RJ, McPherson CA. Factors affecting the rate of breakdown of bacterial protein in rumen fluid. *Br J Nut*. 1987; 58: 313-323.
17. Pinares-Patiño CS, Ulyatt MJ, Waghorn GC, Lassey KR, Barry TN, Holmes CW, Johnson DE. Methane emission by alpaca and sheep fed on lucerne hay or grazed on pastures of perennial ryegrass/white clover or birds foot trefoil. *J Agric Sci*. 2003; 140: 215–226.
18. Woodward SL, Waghorn GC, Laboyre P. Condensed tannins in birdsfoot trefoil (Lotus corniculatus) reduced methane emissions from dairy cow. *Proc NZ Soc Anim Prod*. 2004; 64: 160-164.
19. Puchala R, Min BR, Goetsch AL, Sahlu T. The effect of a condensed tannin- containing forage on methane emission by goats. *J Anim Sci*. 2005; 83: 182-186.
20. Augusti KT. Therapeutic values of onion (Allium cepa) and garlic (Allium sativum). *Indian J Exp Biol*. 1996; 34: 634-40.
21. Pandian Suja R, Anuradha CV, Viswanathan P. Gastroprotective effect of fenugreek seeds (Trigonella foenum- graecum) on experimental gastric ulcer in rats; *J Ethnopharmacol*. 2001; 81 (2002): 393-397.
22. Sahoo T, Tiwari SP, Ravikanth K, Thakur A, Maini S. A new polyherbal formulation for lean meat production in broilers. *Int J Advanced Res*. 2013; 1(9): 70-76.
23. Mane B, Ravikanth K, Thakur A. Lean Meat Production in Broilers with New Polyherbal Formulation AV/LMP/10. *Indian J Applied Res*. 2014; 4(1): 527-530.
24. Rahimil S, Teymouri ZZ, Karimi Torshizi MA, Omidbaigi R, Rokn H. Effect of the Three Herbal Extracts on Growth Performance, Immune System, Blood Factors and Intestinal Selected Bacterial Population in Broiler Chickens. *J Agri Sci and Technol*. 2011; 13: 527-53.
25. Bristow DJ, Holmes DS. Cortisol levels and anxiety-related behaviors in cattle. *Physiol Behav*. 2007; 90: 626-628.
26. Ali A, Birol D, Ramazan A. The effects of fenugreek (*Trigonella foenum graecum l.*) seeds on the performance and some carcass traits of quail (coturnix coturnix japonica) chicks. *The International*. 2011; 54: 115.
27. Elagib HAA, El-Amin WIA, Elamin K M, Malik HE. Effect of Dietary Garlic (*Allium sativum*) Supplementation as Feed Additive on Broiler Performance and Blood Profile. *J Anim Sci Adv*. 2013; 3(2): 58-64.

Table 1. Methane emission estimation *in vivo* on DDMI, DMI basis

Particulars	T_0	T_1
Recovered CH_4volume (L)	46.290[a]	42.310[b]
Recovered CH_4weight (g)	33.064[a]	30.221[b]
Methane emission (L)/kg of DDMI	66.251[a]	58.212[b]
Methane weight emitted (gms)/kg of DDMI	47.322[a]	41.580[b]
Methane emission (L)/kg of DMI	113.579[a]	95.964[b]
Methane weight emitted (gms)/kg of DMI	81.128[a]	68.546[b]

[ab]Values with different superscripts in a rows differ at 5% level of significance

Table 2. Percentage phenolic content of the supplemented concentrate ration

Particulars	T_0	T_1
Total phenolics (%)	5.23[a]	5.47[b]
Non-tannin phenolics (%)	2.23[a]	1.77[b]
Pure tannin phenolics (%)	3.00[a]	3.70[b]
Condensed tannins (CT) (%)	5.34[a]	6.89[b]

[ab]Values with different superscripts in a rows differ at 5% level of significance

Table 3. Plan of nutrition of Deccani sheep

Particulars	T_0	T_1
TDN Intake (g/d)	422.51[a]	453.73[b]
DCP Intake (g/d)	56.55[a]	64.17[b]
DE Intake (MJ/d)	7.80[a]	8.37[b]
ME Intake (MJ/d)	6.39[a]	6.86[b]

[ab]Values with different superscripts in a rows differ at 5% level of significance

Table 4. Dry matter intake and feed conversion efficiency in Deccani sheep

Groups	Initial B.wt. (kg)	Final B.wt. (kg)	DMI (g/day)	DMI (kg/100kg b.wt.)	FCR (kg/kg wt. gain)
T_0	16.45±0.83	20.23±0.83[a]	698.71±12.12[a]	4.25[a]	16.64±1.2[a]
T_1	16.48±0.65	20.95±1.32[b]	726.83±15.18[b]	4.41[b]	14.63±1.23[b]

[ab]Means with different superscripts in a column differ at 5% level of significance

Table 5. Digestibility coefficients of nutrients

Groups	Dry matter	Organic matter	Crude fibre	Crude protein	Ether extract	NFE	NDF	ADF
T_0	58.33[a]	68.80[a]	73.73[a]	55.86[a]	53.14[a]	62.26[a]	67.68[a]	72.19[a]
T_1	60.66[b]	71.15[b]	75.51[b]	57.44[b]	54.99[b]	64.83[b]	70.06[b]	74.19[b]

[ab]Values with different superscripts in a column differ at 5% level of significance

Table 6. Mean body weight (kg) and average daily gain (g/day) (Mean ± SE) at fortnightly intervals in Deccani sheep

Particulars	T_0	T_1
0 Days	16.45±0.83	16.48±0.65
15 Days	16.63±0.76[a]	16.59±0.71[b]
30 Days	16.67±0.98[a]	17.21±0.98[b]
45 Days	17.36±0.82[a]	17.69±1.06[a]
60 Days	18.18±0.91[a]	18.39±1.09[a]
75 Days	19.21±0.82[a]	19.82±1.23[b]
90 Days	20.23±0.83[a]	20.95±1.32[b]
ADG (gms/d)	42.07±5.69[a]	49.64±7.74[b]

[ab]Means with different superscripts in the same row differ at 5% level of significance

Table 7. Nitrogen balance of control and treatment groups on experimental basis

Particulars	T_0	T_1
Nitrogen intake (g/day)	9.05±0.76[a]	10.27±0.34[b]
Faecal nitrogen (g/day)	1.77±0.01	1.78±0.03
Digested nitrogen (g/day)	7.28±0.56[a]	8.49±0.68[b]
Urinary nitrogen (g/day)	3.95±0.02[a]	3.55±0.03[b]
Nitrogen balance (g/day)	3.33±0.12[a]	4.94±0.21[b]

[ab]Values with different superscripts in a rows differ at 5% level of significance

Table 8. Blood cortisol (ng/mL) level in both groups of Deccani sheep at different time intervals

Days	T_0	T_1
0 day	40.01±1.69[a]	42.22±2.38[b]
45 day	37.08±3.20[a]	27.25±1.14[b]
90 day	22.56±1.43[a]	7.71±0.94[b]
Overall	33.12±2.02[a]	25.73±1.56[b]

[ab]Values with different superscripts in a rows differ at 5% level of significance

Table 9. Carcass quality traits (Mean±SEM) in control and treatment groups

Body parts	T_0	T_1
All 4 feet (kg)	0.50±0.04[a]	0.48±0.02[a]
Head weight (kg)	1.46±0.24[a]	1.22±0.02[a]
Intestine with content (kg)	1.30±0.36[a]	1.19±0.01[b]
Stomach with content (kg)	3.44±0.86[a]	3.10±0.36[a]
Pluck (kg)	0.71±0.04[a]	0.68±0.04[b]
Separable fat weight (kg)	0.11±0.01[a]	0.22±0.12[b]
Skin (kg)	2.70±0.78[a]	2.03±0.07[a]
Blood weight (kg)	0.57±0.05[a]	0.65±0.01[b]
Live weight (Kg)	19.10±3.90[a]	18.25±0.75[a]
Dressed carcass weight (kg)	8.46±1.96[a]	8.91±0.91[b]
Dressing (%)	44.1±1.02[a]	49.3±3.01[b]

[ab]Values with different superscripts in a rows differ at 5% level of significance

Figure 1. Animal in the closed respiratory chamber for methane emission estimation