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EFFECT OF BHRINGRAJ (*Eclipta alba*) HERB ON *IN VITRO* FERMENTATION CHARACTERISTICS OF BAJRA STRAW AND BAJRA STRAW-BASED COMPLETE FEED

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ABSTRACT

An experiment was conducted to assess the effect of Bhringraj (*Eclipta alba*) herb supplementation to bajra straw @ 0, 1, 2, 3 and 4% kg^{-1} dry matter substrate on degradability of dry matter (DM), organic matter (OM) and neutral detergent fibre (NDF) and total gas production (TGP) using goat rumen liquor. Significant improvement in nutrient degradability of DM, OM and NDF in bajra straw and TGP production was observed due to Bhringraj supplementation with maximum values at 3% kg^{-1} DM. Another *in vitro* trial was performed to evaluate the effect of 3% Bhringraj kg^{-1} DM on rumen fermentation characteristics in complete feed having bajra straw and concentrate mixture in 60:40 ratio. The treatment significantly improved nutrient degradability (DM, OM and NDF), TGP, total rumen nitrogen and TVFA in comparison to control. Addition of Bhringraj in complete feed did not alter rumen pH, however there was significant decrease in rumen ammonia-nitrogen in herb supplemented complete feed.

Keywords: Bajra straw, complete feed, *Eclipta alba*, gas production, *in vitro* degradability, rumen fermentation

INTRODUCTION

The ever increasing number of consumers demanding healthy and natural foods have pushed organic livestock farming in the present era. This system of livestock farming is environmental friendly, sustains animals in good health with high welfare standards and prohibits routine use of growth promoters, animal offals and antibiotics or any other chemicals and additives in the livestock ration. It is, therefore, imperative to explore alternative natural feed additives to improve animal productivity. The use of herbs in livestock ration as feed additives is receiving increasing attention as it is considered safe without any side effect as well as enhances nutrient utilization and productivity in animals. The beneficial effects of herbs in farm animals may arise from activation of feed intake and secretion of digestive enzymes, improved feed efficiency, reduced incidence of digestive disturbance, stimulation of digestion, methane inhibition, improved reproductive parameters and productive performance, immuno-stimulation, antibacterial, coccidiostatic, anthelmentic, antiviral or antioxidative characteristics (Uegaki *et al*., 2001). Herbs and/or herbal preparations stimulate the endocrine system and enhance the intermediate nutrient metabolism, thereby enhances the nutritional status of animal by maintaining the blood biochemical constituents and enzyme profile concerned with metabolic activities. The extent to which level the herbs can be incorporated in livestock ration to increase nutrient intake and utilization as well as animal performance has not been well established. Therefore, the present investigation was undertaken to study the effect of herb Bhringraj

(*Eclipta alba*) at different levels on *in vitro* nutrient degradability characteristics and total gas production of bajra straw and fermentation pattern in bajra straw-based complete feed using rumen liquor of goats.

MATERIALS AND METHODS

The samples of bajra straw, complete feed and Bhringraj herb were oven-dried, ground and passed through 1 mm sieve in a Willy mill and stored in plastic bottles for further use. The proximate constituents of bajra straw, Bhringraj and complete feed was done as per AOAC (1999) and fibre fractions as per VanSoest *et al.* (1991). The modified *in vitro* procedure of Tilley and Terry (1963) was used to assess *in vitro* dry matter degradability (IVDMD), *in vitro* organic matter degradability (IVOMD) and *in vitro* nutrient detergent fibre degradability (IVNDFD) at 48 and 72 hr post-incubation and total gas production at 24 hr post-incubation. Three adult bucks were given 20 days adoption time to complete feed containing bajra straw at 60% level to meet nutrient requirements as per ICAR (1998). Rumen fluid was collected from these bucks before morning feeding (Solvia and Hess, 2007) by perforated tubing device under negative pressure and squeezed through four layers of muslin cloth to get inoculums (SRL), then transferred to pre-warmed flask (39ºC) and flushed with CO_2. SRL (10 ml) was dispensed into pre-warmed (39ºC) fermentation vessels (Jakhmola *et al.*, 2010) containing 500 mg finely ground bajra straw (1 mm) without or with 1, 2, 3and 4% herb kg^{-1} substrate DM + 40 ml buffer and flushed with oxygen free CO_2. The experiment was conducted in completely randomized block design and each treatment run in triplicate with negative controls (SRL + buffer alone). The controls were used to correct for fermentation residues resulting directly from SRL. Each vessel cork fitted with control value was kept in incubator at 39ºC for 48 hr. *In vitro* gas production was measured periodically upto 24 hr incubation by Doctor's syringe (Jakhmola *et al.*, 2010). At the end of 48 hr incubation two drops of saturated $HgCl_2$ was added in each vessel to stop microbial activity. Contents of each vessel were transferred to 1 L spout-less beakers. The vessels were thoroughly washed with neutral detergent solution and final volume made to 150 ml. The contents were refluxed for 1 hr at 100ºC, filtered and washed through pre-weighed Gooch crucible (Grade 1, 50 ml capacity). This undigested residue (NDF) was oven-dried at 100ºC for 24 hr, cooled in desiccator and weighed. Loss in DM and NDF was digested dry matter and digested NDF. The crucibles containing residue were ignited in muffle furnace at 500ºC and the ash left in crucible after ignition was subtracted from residual dry matter to get the organic matter content. For *in vitro* studies at 72 hr, incubation of samples was done in same way as for 48 hr incubation. The reaction was stopped by adding 2 ml of 6 N HCl and 0.1 g pepsin powder (1:3000) to each vessel at the end of 48 hr incubation. Then the vessels were incubated for another 24 hr and procedure repeated as for 48 hr incubation, except the addition of $HgCl_2$ at 48 hr.

The optimum level of herb, chosen from experiment I, was mixed with finely ground (1 mm) complete feed containing bajra straw and concentrate mixture in 60:40 ratio. The concentrate mixture contained barley 04, deoiled rice bran 04, groundnut cake 15, gaur korma 14, mineral mixture 02 and salt 01 parts. The experimental procedures for degradability of nutrients in complete feed were similar to experiment I. The pH in rumen liquor was determined immediately after the termination of incubation at 48 hr using portable digital pH meter. Total volatile fatty acids was determined as per method of Barnett and Reid (1957) using Markham still distillation apparatus, total rumen nitrogen as per AOAC (1999) and ammonia nitrogen by spectrophotometer method (Chaney and Marbach, 1962). The data were analyzed statistically as per Snedecor and Cochran (1994) to draw the inference.

RESULTS AND DISCUSSION

The herb *Eclipta alba* contained 9.8% crude protein, 0.91% ether extract, 11.5% crude fibre, 68.5% N-free extract, 52.3% neutral detergent fibre, 46.5% acid detergent fibre, 5.87% hemicellulose and 5.86% cellulose (Table 1). The chemical composition of Bhringraj was comparable with Samal *et al.* (2009) and Manju (2010). The chemical composition of bajra straw is comparable with Dhuria *et al.* (2007).

Table 1: Chemical composition bajra straw, complete feed and Bhringraj

Chemical components (% DM)	Bajra straw	Complete feed	Bhringraj (*E. alba*)
Dry matter	93.05	94.03	96.11
Organic matter	91.02	89.30	91.65
Crude protein	4.60	16.42	9.80
Ether extract	1.24	2.41	0.91
Crude fibre	34.21	22.53	11.50
Nitrogen free extract	50.97	47.94	68.51
Neutral detergent fibre	78.51	58.11	52.32
Acid detergent fibre	54.41	37.41	46.45
Hemicellulose	24.10	20.70	5.87
Cellulose	28.51	24.35	5.86

The IVDMD and IVOMD of bajra straw supplemented with Bhringraj was significantly higher as compared to control at 2, 3 and 4% level at both 48 and 72 hr incubation (Table 2). Bhringraj supplementation did not significantly influence the degradability at 1%. Maximum IVDMD and IVOMD were noticed at 3% supplementation though the differences between 3 and 4% were at par. Bhringraj supplementation improved DM degradability at 48 and 72 hr incubation by 3.30 and 4.72, 5.98 and 7.11, 10.14 and 10.09, 9.66 and 8.83% over control at 1, 2, 3 and 4% supplementation level, respectively. A trend similar to IVDMD was observed in IVOMD. The respective improvement in OM degradability for 48 and 72 hr incubation was 3.76 and 4.08, 15.75 and 10.29, 22.86 and 15.32, 21.52 and 12.62% over control at 1, 2, 3 and 4% Bhringraj supplementation, respectively. Addition of Bhringraj herb did not affect degradability of IVNDFD to significant level at 48 hr incubation, irrespective of level of supplementation; whereas a significant effect was noticed at 72 hr incubation. The respective improvement over control in IVNDFD for 48 and 72 hr incubation was 2.13 and 1.31, 7.87 and 9.90, 13.20 and 15.39 and 10.09 and 10.50% at 1, 2, 3 and 4% Bhringraj, respectively. A linear increase in IVDMD, IVOMD and IVNDFD at 48 and 72 h incubation with increase in the level of supplementation of Bhringraj upto 3% DM was observed and thereafter inclusion of Bhringraj did not show any additional improvement on IVDMD, IVOMD and IVNDFD. The results suggest that the addition of herb might have increased the population of fibre degrading bacteria and/or their activity (Das, 1992; Manjunatha, 1998). These findings corroborate with those of Bakshi *et al.* (2004) who reported that *in vitro* digestibility of nutrients and availability of metabolizable energy improved with herbal feed additives using berseem and wheat straw as substrate. The findings are in line with Gupta *et al*. (2005) and Kumar *et al*. (2006) who reported that supplementation of herbs at 5% level increased IVDMD of paddy straw. Sardar *et al*. (1998) observed that a mixture of herb supplementation (50 mg g^{-1}) improved IVDMD of oat fodder, concentrate mixture and oat fodder + concentrate mixture.

Table 2: *In vitro* DM, OM and NDF degradability, and total gas production in bajra straw

Attribute	Incubation (hr)	Bhringraj supplementation (%)					SEM
		0	1	2	3	4	
IVDMD (%DM)	48	41.51^{a}	42.88^{a}	47.49^{b}	51.65^{c}	51.17^{c}	0.934
	72	47.83^{a}	50.09^{a}	54.94^{b}	57.92^{b}	56.66^{b}	1.069
IVOMD (%DM)	48	43.53^{a}	45.17^{a}	50.31^{b}	53.48^{c}	52.90^{c}	0.726
	72	50.69^{a}	52.76^{a}	55.90^{b}	58.46^{c}	57.09^{bc}	0.769
IVNDFD (%DM)	48	29.62^{a}	30.25^{a}	31.95^{a}	33.53^{a}	32.51^{a}	1.119
	72	33.52^{a}	33.96^{a}	36.84^{b}	38.68^{b}	37.04^{b}	0.816
IVTGP (ml 500 mg^{-1})	24	28.10^{a}	33.43^{b}	35.92^{b}	36.58^{b}	33.75^{b}	1.194

Note: Means superscripted with different letters in a row differ significantly ($P<0.01$)

The improvement in IVTGP in bajra straw supplemented with 1, 2, 3, and 4% of herb was 18.97, 27.83, 30.18 and 20.11% over control at 24 hr incubation. The results fall in line with the observations of Bakshi *et al.* (2004) who reported increased net gas production in berseem and wheat straw on supplementation of different herbs. The increase in total gas production appeared to be associated with

higher DM, OM and NDF degradability in substrate due to increase in bacterial population, activity and/or rate of fermentation.

Digestibility of nutrients in *in-vitro* system was more pronounced when complete feed containing bajra straw and concentrate mixture at 60: 40 ratio was used as substrate showing significant effect of herb supplementation on degradability of DM, OM and NDF and total gas production (Table 3) when compared with unsupplemented complete feed at 48 hr incubation. IVDMD, IVOMD, IVNDFD and TGP increased by 20.94, 20.93, 7.66 and 18.71%, respectively, over unsupplemented complete feed. The results corroborate with those of Manju, (2010), who reported increased degradability of nutrients and total gas production when *Eclipta alba* was added in complete feed. Ishtiyak *et al*. (2010) also observed increase in IVDMD and IVOMD on supplementation of different herbs in complete feed at 2 and 3% level in rumen fluid of goats. Similar findings have been reported by Sardar *et al.* (1998) and Wadhwa and Bakshi (2006). Bhringraj supplementation did not alter pH of *in vitro* rumen fluid of goats which were within normal range of rumen pH (6.5-7.0) indicating no adverse effect of herb addition on rumen environmental conditions. The results are in agreement with Ishtiyak *et al*. (2010). Total nitrogen and total volatile fatty acid concentration was significantly higher (93.99 mg dl^{-1} and 84.91 m Eq L^{-1}) in herb supplemented complete feed as compared to control (92.16 mg dl^{-1} and 81.22 m Eq L^{-1}), which indicated stimulatory effects of herb on fibre degrading microorganisms. It appears that more energy may be available due to herb supplementation. Contrarily, there was significant decrease in rumen ammonia-nitrogen in herb supplemented complete feed as compared to unsupplemented one, indicating higher incorporation of ammonia-nitrogen into microbial protein (Sardar *et al*., 1998; Wanapat *et al.*, 2008). The results recorded are authenticated by Manju (2010) and Wanapat *et al.* (2008) in *in-vivo* experiments with sheep and cattle, respectively.

Table 3: *In vitro* degradability of nutrients and rumen metabolites in complete feed

Parameter	Unsupplemented	Bhringraj supplemented	SEM
IVDMD (% DM)	51.78^a	62.62^b	0.983
IVOMD (% DM)	53.27^a	64.42^b	0.814
IVNDFD (% DM)	45.01^a	48.46^b	0.747
IVTGP (ml 500 mg^{-1})	53.67^a	63.71^b	0.769
pH	6.84^a	6.82^a	0.021
TVFA (m Eq L^{-1})	81.22^a	84.91^b	0.121
Total N (mg/dl)	92.16^a	93.99^b	0.062
Ammonia N (mg/dl)	32.94^a	31.01^b	0.171

Note: Means superscripted with different letters in a row differ significantly ($P<0.01$)

Conclusion: The study revealed a significant improvement in digestibility of nutrients and rumen fermentation in *in-vitro* rumen fluid of goats as an effect of supplementation of Bhringraj (*Eclipta alba*) at 3 % level of DM. However, *in vivo* trial in goats is required before advocating these findings to goat raisers.

REFERENCES

AOAC. 1999. *Official Methods of Analysis*, Association of Official Analytical Chemists (18th edn.), Washington, DC, USA.

Bakshi, M.P.S., Rani, N., Wadhwa, M. and Kaushal, S. 2004. Impact of herbal feed additives on the degradability of feed stuffs *in vitro. Indian Journal of Animal Nutr*ition, **21**: 249 – 253.

Barnett, A.J.G. and Reid, R.L. 1957. Studies on the production of VFA from grass by rumen liquor in artificial rumen: The VFA production from fresh grass. *Journal of Agricultural Science,* **48**: 315.

Chaney, A.L. and Marbach, E.P. 1962. Modified reagents for determination of urea and ammonia. *Clinical Chemistry*, **8**: 130-133

Das, S.N. 1992. A study of the effect of BH-strong on ruminal microflora for improved digestion in cattle. *Indian Journal of Indigenous Medicine,* **9**: 31–37.

Dhuria, R.K., Purohit, G.R. and Sharma, T. 2007. Evaluation of bajra straw based complete feed in sheep. *Indian Journal of Animal Nutrition*, **24**: 209-211.

Gupta, N., Kumar, A. and Tiwari, D.P. 2005. Effect of herbs as feed additive on nutrient utilization and growth in crossbred heifers fed paddy straw based ration. *Indian Journal of Animal Science*, **75**: 52–55.

Haliwell, B.R., Loeliger, A.J. and Aruoma, O.I. 1995. The characterizatioin of antioxidants. *Food and Chemical Toxicology*, **33**: 601–607.

ICAR. 1998. *Nutrient Requirement of Livestock and Poultry*. (2nd revised ed.) Indian Council of Agricultural Research, New Delhi, India.

Ishtiyak, A.M., Kumar, R., Sharma, R.K. and Barman, K. 2010. Effect of herbs on *in vitro* digestibility of feed with rumen liquor of goat. *Indian Journal of Veterinary Research*, **19**: 13-18.

Jakhmola, R.C., Raghuvanshi, S.K.S., Singh, N. and Tripathi, M.K. 2010. *Fermentation Vessel for Conducting Gas Production Studies (in vitro)*. Patent No. 6642/ PQ-DEL/2010 dt. 06/09/2010. ICAR, New Delhi, India.

Kumar, A., Gupta, N. and Tiwari, D.P. 2006. Effect of herbs as feed additive on *in vitro* and *in sacco* dry matter digestibility of paddy straw. *Indian Journal of Animal Sci*ence, **76**: 847–850.

Manju, 2010. *Effect of Feeding Herbs as Feed Additive on Nutrient Utilization Efficiency in Sheep.* M.V.Sc. thesis submitted to Swami Keshwanand Rajasthan Agricultural University Bikaner, Rajasthan, India.

Manjunatha, B.P. 1998. Effect of herbal biostimulator on ruminant microbial digestion. *Pashudhan* **13**(4): 4.

Samal, L., Ingale, S.L., Kankoriya, S., Ballyan, S., Ravikanth, K. and Pattanaik, A.K. 2009. An investigation on the nutritional composition of some herbal feed additives of known functional properties. p. 205. In: *Proceedings of Animal Nutrition Association World Conference*, Feb., 14-17, New Delhi, India.

Sardar, P., Kewalramani, N. and Kaur, H. 1998. Effect of *Rumbion bolus* supplementation on rumen fermentation and IVDMD. *Indian Journal of Dairy Sci*ence, **51**: 40-43.

Snedecor, G.W. and Cochran, W.C. 1994. *Statistical Methods* (8th edn.). Oxford and IBH Publishing Co., New Delhi, India.

Solvia, C.R. and Hess, H.D. 2007. Measuring methane emission of ruminants by *in vitro* and *in vivo* technique. pp. 15-31. In: *Measuring Methane Production from Ruminants* (Eds. H.P.S. Makkar and P.E. Vercoe), IAEA, Vienna, Austria.

Tilley, J.M.A. and Terry, R.A. 1963. A two-stage technique for the *in vitro* digestion of forage crops. *Journal of* British *Grassland Society*, **18**: 104-111.

Uegaki, R., Ando, S., Ishida, M., Takada, D., Shinokura, K. and Kohchi, Y. 2001. Antioxidant activity of milk from cows fed herbs. *Nippon Nogeikagaku Kaishi*, **75**: 669 – 671.

Vansoest, P.J, Robertson J.B and Lewis B.A. 1991. Methods of dietary fiber, neutral detergent fiber and non-starch polysaccharides in relation to animal nutrition. *Journal of Dairy Science*, **74**: 3583-3597.

Wadhwa, M. and Bakshi, M.P.S. 2006. Herbal feed additives - Impact on the rumen environment in buffaloes. *Indian Journal of Animal Nutrition*, **23**: 102-109.

Wanapat, M., Cherdthong, A., Pakdee, P. and Wanapat, S. 2008. Manipulation of rumen ecology by dietary lemongrass (*Cymbopogon citrates Stapf.*) powder supplementation. *Journal of Animal Science*, **86**: 3197-3503.