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Evaluation of rumen fermentation parameters and hematological profile in Murrah buffalo calves maintained on total mixed ration containing agricultural byproducts

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Abstract

A study was conducted to assess the rumen fermentation pattern and nutrient utilization in Murrah buffalo calves fed with different total mixed rations (TMR). Six Murrah buffalo calves were selected as uniformly as possible with regard to age (8-12 months), sex and body weight from University Livestock Farm and Fodder Research and Development Scheme, Mannuthy and formed treatment group. Three feeding trials for a period of one month each using TMR's containing concentrate mixture I, (BIS-2012) [1] with conventional feed ingredients in trial-I, concentrate mixture II containing turmeric waste and concentrate mixture III containing tapioca starch waste in trial II and III, respectively. The green grass was used as roughage in all the three trials. The rumen fermentation profile, nutrient digestibility, hematological profile and microbial protein production were studied. The results revealed that Murrah buffaloes, fed with TMRs containing turmeric waste (trial-II) and tapioca starch waste (trial-III) did not show any significant difference in rumen fermentation parameters compared to groups fed with TMR-I (control). The volatile fatty acid profile showed increased concentration of propionic acid in animals fed with TMR-III and the acetate: propionate ratio in those fed with TMR-II. Hematological parameters were well within the normal range and were not significant among animals of different groups. It can be concluded that the ayurvedic pharmaceutical byproducts can be incorporated in the total mixed ration without affecting the rumen fermentation pattern and hematological profile in Murrah buffalo calves. The volatile fatty acid profile were improved in animals fed with TMR containing agricultural byproducts compared to the conventional TMR.

Keywords: Total mixed ration, murrah buffaloes, turmeric waste, tapioca starch waste, rumen fermentation profile, volatile fatty acid profile

Introduction

The common feeding regimen followed by farmers from the good old days was to feed their livestock with concentrate and roughage as separate allowances. The limited control over the proportion of concentrate and roughage intake as well as the selection done by the livestock made this method of feeding inadequate. Total mixed ration (TMR) feeding helps in stabilization of rumen fermentation and ensures better ammonia utilization and help in incorporating poor quality roughage and unconventional feedstuffs (Sato *et al.*, 1987) [8]. TMR feeding will also ensure the sufficient concentrate: roughage in lactating animals, since selection among feed ingredients is not possible. In Kerala there is a huge gap between feed and fodder requirement and availability (Economic Review., 2017) [3]. This tempted the scientist to tryout newer low cost but nutritionally rich feed resources as alternate feeds. The agricultural crop residues, fruit and vegetable processing industry and ayurvedic pharmaceutical industry byproducts contributes the major share of these unconventional feeds. The vegetable wastes such as cauliflower leaves, cabbage leaves, pea pods and the pea vines are used as an efficient replacement for roughages. The dhanvantharam oil residues, tapioca starch waste, coconut poonac and Ksheerabala residues can be used as an effective replacement for conventional concentrates in livestock ration (Rani *et al.*, 2016) [7]. The agricultural byproducts which are available in large quantities can be used in TMR as ingredients for livestock feeding. The burning of crop residues will result in environmental pollution and huge economic loss, which otherwise can be used for feeding livestock and also

for bio fuel production. However the influence of unconventional feed inclusion on rumen fermentation pattern and nutrient utilization is not much studied in Murrah buffaloes. Hence, the study has been taken up to evaluate the rumen fermentation pattern of Murrah buffalo calves, maintained on the standard and selected TMRs containing unconventional feeds.

Materials and methods

The research work was conducted at University Livestock Farm and Fodder Research and Development Scheme, The Department of Animal Nutrition, College of Veterinary and Animal Sciences, Mannuthy, Kerala Veterinary & Animal Sciences University. Six calves of Murrah buffalo were selected as uniformly as possible with regard to age (between 9-12 months), sex and body weight from University Livestock Farm and Fodder Research and Development Scheme, Mannuthy. These animals were maintained under uniform management conditions prevailing in the farm. All the animals were dewormed for controlling endoparasites. A feeding trial for a period of one month using TMR containing concentrate mixture I (BIS-2012) [1] with conventional feed ingredients and green grass as roughage was carried out. TMR II containing turmeric waste and TMR III containing tapioca starch waste replacing conventional feed ingredients and green grass as roughage was used for the two subsequent feeding trials for one month each. All the animals were fed according to ICAR (2013) [4] recommendations and the concentrate: roughage ratio maintained was 50: 50. The ingredient composition of the compound feed mixtures used

in three TMR's was presented in Table 1 and chemical composition of rations fed to Murrah buffaloes in three trials were presented in Table-2.

Rumen Fermentation Parameters

Rumen liquor was collected from all the six animals at the beginning and end of the feeding trials using a stomach tube after three hours of feeding into a pre warmed thermos- flask maintained in anaerobic condition at 39°C and brought to the laboratory and subjected to further analysis. Rumen fermentation parameters such as rumen pH was measured using a digital pH meter, volatile fatty acids using gas chromatograph, total nitrogen (Kjeldahl method), trichloro acetic acid nitrogen (Kjeldahl method) and ammonia nitrogen using the colorimetric method described by (Beecher and Whittan., 1970) [2].

Hematological Parameters

Blood samples were collected at the beginning (0 day) and end of each feeding trial from all the animals to estimate blood urea nitrogen (modified Berthlot method), haemoglobin, volume of packed red cells (VPRC) using haematology analyzer, Glucose (GOP-PAP method), Creatinine (modified Jaffe method) using standard kits from Agappe Diagnostics, Cochin, Kerala. Data gathered on the various parameters, in all the three groups were analyzed statistically as per Snedecor and Cochran (1994) [10] by analysis of variance (ANOVA) technique, using the software, statistical package for social sciences (SPSS) version 24.0.

Table 1: The ingredient composition of the concentrate mixture used for three trials

Ingredients	Concentrate - I	Concentrate - II	Concentrate - III
Maize	31.5	28	32
Bajra	9.5	7	2
Soya bean meal	22	22	24
Wheat bran	9.5	11.5	2
De oiled rice bran	23	22	30.5
Turmeric waste	-	5	-
Tapioca starch waste	-	-	5
Mineral mixture	3	3	3
Salt	1.5	1.5	1.5
Total	100	100	100

Table 2: Chemical Composition of rations fed to Murrah Buffaloes, % in three trials

Nutrient	Green grass	Concentrate-I	Concentrate- II	Concentrate- III
Dry matter*	20.25	91.03	91.00	90.62
Crude protein*	11.60	20.04	20.03	19.62
Ether extract*	1.48	6.39	1.80	1.33
Crude fibre*	35.57	2.09	1.53	4.96
Neutral Detergent Fiber*	59.04	26.81	43.57	40.50
Acid detergent fiber*	38.20	17.20	22.10	17.91
Total ash*	12.63	8.62	8.13	7.96
Acid insoluble ash*	3.55	1.05	1.46	1.05
Nitrogen free extract*	38.72	60.88	66.51	66.13
Organic matter*	87.37	91.38	91.87	92.04

*Values expressed on the DM basis, average of six values

Results and Discussion

Data pertaining to rumen fermentation parameters, the volatile fatty acid profile and hematological profile of Murrah buffaloes fed with TMR containing conventional ingredients and agricultural byproducts are presented in table-3, 4 and 5

Rumen fermentation parameters

The results revealed that the rumen pH, rumen ammonia nitrogen (mg/100 ml), trichloro acetic acid precipitable nitrogen(mg/100 ml), total nitrogen (mg/100 ml) were 6.8 ± 0.04 , 6.8 ± 0.03 and 6.82 ± 0.03 , 15.22 ± 0.73 , 15 ± 0.58 and 14.84 ± 0.53 , 41.42 ± 1.4 , 42.25 ± 0.73 and 41.92 ± 0.82 , 70.33 ± 1.84 , 73.33 ± 3 and 75.67 ± 2.39 respectively for

animals fed TMR-I (control), TMR-II and TMR-III. The statistical analysis revealed that there was no significant difference among values recorded for parameters such as pH, rumen ammonia nitrogen, TCA precipitable nitrogen and total nitrogen. Thakur *et al.*, (2005) [11] reported that TMRs did not have any significant effect on rumen fermentation parameters in male Murrah buffalo calves of 8-9 months old fed with TMR1- consisted of concentrate + green maize fodder + wheat straw in ratio 50:25:25, TMR2 and TMR3 contained same ingredients as in TMR I but in concentrate maize and barley were replaced with wheat, ground nut cake with mustard cake and urea. Poonooru *et al.*, (2015) [6] reported an increase in concentration of the rumen ammonia nitrogen and total nitrogen in buffalo bulls fed TMR supplemented with exogenous fibrolytic enzymes (EFE) and live yeast culture T1 (TMR at 70:30 roughage and concentrate), T2 (T1 + 15 g/animal/dry EFE, T3 = T1 + 10 g live yeast/animal/day, T4 = T1 + 15 g EFE + 10 g live yeast. The results revealed that animals fed T4, T3, T2 had significantly higher ammonia nitrogen, TCA nitrogen and total nitrogen compared to those fed T1. Further they reported that there was no significant effect on rumen pH in all treatment groups.

Rumen Volatile Fatty Acid Profile in Murrah Buffaloes

The data revealed that acetic acid concentration (mmol/L) in animals maintained on TMR I, II and III during the Trail I, Trial II and Trial III was 54.03 ± 1.59 , 57.03 ± 1.28 and 55.36 ± 1.21 mmol/L, propionic acid concentration were 22.23 ± 0.95 , 21.62 ± 0.62 and 26.46 ± 0.33 mmol/L respectively in animals maintained during Trial I, Trial II and Trial III while rumen butyric acid concentration was 2.11 ± 0.26 , 2.96 ± 0.24 and 2.90 ± 0.20 mmol/L respectively. Total volatile fatty acid concentration in animals maintained on TMR I,II and III during Trial-I, Trial II and Trial III was 78.37 ± 2.11 , 81.6 ± 0.98 and 84.72 ± 1.18 mmol/L, respectively. The acetate: propionate ratio was 2.45 ± 0.11 , 2.65 ± 0.12 and 2.09 ± 0.06

respectively. The statistical analysis revealed that propionic acid concentration was higher ($P < 0.05$) in animals maintained on TMR-III containing tapioca starch waste during Trial-III. Higher propionic acid concentration was due to the inclusion of tapioca starch waste in TMR fed to animals in Trial III. Acetate: propionate ratio in animals maintained on Trial I and II was significantly higher ($P < 0.05$) than animals in trial -III and there was no significant difference among values recorded for acetic acid, butyric acid and the total volatile fatty acid for animals maintained on trial-I, II and III. Wadhwa and Bakshi (2006) [12] reported that total volatile fatty acid concentration was higher in TMR fed groups than control group fed concentrate and roughage separately. Poonooru *et al.* (2015) [6] reported that TVFA was lower in TMR fed animals compared to those supplemented with exogenous fibrolytic enzyme or live yeast culture (81.56 vs 89.46 mmol/L).

The haematological profile of Murrah Buffalo

The parameters assessed were blood glucose (mg/100 ml), blood urea nitrogen (mg/100 ml), creatinine (mg/100 ml), haemoglobin (g/100 ml), red blood cells ($10^6/\mu\text{l}$), the volume of packed red cells (VPRC%). The data revealed that blood glucose level in animals maintained during Trial-I, II and III periods were 67.78 ± 2.91 , 67.67 ± 2.4 and 69.67 ± 2.11 while BUN concentrations were 28.41 ± 1.78 , 27.48 ± 1.7 and 23.48 ± 0.83 , respectively. The Serum creatinine concentration in animals maintained during Trial-I, Trial II and Trial III periods were 9.9 ± 0.65 , 10 ± 0.62 and 10.5 ± 0.17 and RBC concentration was 6.4 ± 0.36 , 6.48 ± 0.35 and 6.48 ± 0.34 , respectively. The serum VPRC% was 31.77 ± 1.31 , 31.55 ± 1.05 and 33.22 ± 0.61 , respectively for animals maintained during these periods. On statistical analysis it was revealed that there is no significant difference for parameters among animals of the different groups. All the values were in the normal range (Kaneko *et al.*, 2008) [5].

Table 3: Rumen fermentation* parameters in Murrah buffaloes maintained on TMR-I, II and III

Parameters	Trial -I (control)	Trial-II	Trial-III	f-value	p-value
pH	6.8 ± 0.04	6.8 ± 0.03	6.82 ± 0.03	0.294 ^{ns}	0.751
Ammonia nitrogen(mg/100ml)	15.22 ± 0.73	15 ± 0.58	14.84 ± 0.53	0.752 ^{ns}	0.496
TCA precipitable Nitrogen(mg/100ml)	41.42 ± 1.4	42.25 ± 0.73	41.92 ± 0.82	0.315 ^{ns}	0.737
Total Nitrogen (mg/100ml)	70.33 ± 1.84	73.33 ± 3	75.67 ± 2.39	0.550 ^{ns}	0.499

*Mean values are based on six replicates; ns- non significant

Table 4: Rumen volatile fatty acid* profile of Murrah buffaloes maintained on TMR-I, II and III

Parameters	Trial -I (control)	Trial-II	Trial-III	f- value	p- value
Acetic acid (mmol/L)	54.03 ± 1.59	57.03 ± 1.28	55.36 ± 1.21	1.815 ^{ns}	0.213
Propionic acid (mmol/L)	$22.23^b \pm 0.95$	$21.62^b \pm 0.62$	$26.46^a \pm 0.33$	12.808 ^{**}	0.002
Butyric acid (mmol/L)	2.11 ± 0.26	2.96 ± 0.24	2.90 ± 0.20	4.823 ^{ns}	0.079
Total volatile fatty acids (mmol/L)	78.37 ± 2.11	81.6 ± 0.98	84.82 ± 1.18	1.942 ^{ns}	0.222
Acetate: propionate ratio	$2.45^a \pm 0.11$	$2.65^a \pm 0.12$	$2.09^b \pm 0.06$	12.533 ^{**}	0.002

*Significant at 0.05 level; ** significant at 0.01 level; ns- non significant; mean values are based on six replicates; means having different superscripts are significantly different ($p < 0.05$) within a row

Table 5: Hematological profile* of Murrah buffaloes maintained on TMR-I, II and III

Parameters	Trial -I (control)	Trial-II	Trial-III	f- value	p- value
Blood glucose (mg/100ml)	67.78 ± 2.91	67.67 ± 2.4	69.67 ± 2.11	1.345 ^{ns}	0.304
Blood urea nitrogen (mg/100ml)	28.41 ± 1.78	27.48 ± 1.7	23.48 ± 0.83	4.639 ^{ns}	0.077
Creatinine(mg/100ml)	0.92 ± 0.1	0.96 ± 0.07	1.46 ± 0.21	5.114 ^{ns}	0.071
Haemoglobin (g/100ml)	9.9 ± 0.65	10 ± 0.62	10.5 ± 0.17	0.661 ^{ns}	0.538
Red blood cells($10^6/\mu\text{l}$)	6.4 ± 0.36	6.48 ± 0.35	6.48 ± 0.35	5.016 ^{ns}	0.075
Volume of packed red blood cells (%)	31.77 ± 1.31	31.55 ± 1.05	33.22 ± 0.61	1.491 ^{ns}	0.277

NS- non significant; mean values are based on six replicates; means having different superscripts are significantly different ($p < 0.05$) within a row

Conclusion

The results revealed that there were no significant difference among the animals maintained on Trial-I (control), Trial II and Trial III for parameters such as pH, rumen ammonia nitrogen, TCA nitrogen and total nitrogen. The statistical analysis revealed that propionic acid concentration was higher ($P < 0.05$) in animals maintained on Trial III i.e., Murrah buffaloes fed with tapioca starch waste containing TMR. The higher propionic acid concentration was due to the inclusion of tapioca starch waste in TMR. The acetate: propionate ratio in animals maintained on Trial I and trial II was higher than animals fed with TMR containing tapioca starch waste. The hematological parameters among animals on all three trials did not show any significant difference and all the values were in the normal range. It can be concluded that agricultural byproducts such as turmeric waste and tapioca starch waste can be incorporated in total mixed ration without affecting the rumen fermentation pattern and hematological profile in Murrah buffalo calves and the volatile fatty acid profile were improved in animals fed with TMR containing agricultural byproducts compared to the conventional TMR.

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