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Fermentability of complete feed supplemented with different level of buffalo rumen contents as probiotic in balibul local sheep

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Abstract. The aim of the research was to study the effect of complete feed contained different level of buffalo rumen content as probiotic on feed fermentability of Balibul local sheep. Materials used in this study were 16 heads male local sheep under 5 months of age (Balibul), average body weight 11.58 ± 0.77 kg. Experimental design used was completely randomized design (CRD), consisting 4 treatments and 4 replications. Treatments used were T0 (complete feed without buffalo rumen content supplementation), T1 (complete feed supplemented with 5% buffalo rumen content), T2 (complete feed supplemented with 10% buffalo rumen content) and T3 (complete feed supplemented with 15% buffalo rumen content). Parameters observed were VFA, NH₃ and total protein production. Collected data were tested statistically using analysis of variance at 95% confidence interval. Result of the research showed that complete feed supplemented with different level of buffalo rumen content had significant difference ($P < 0.05$) on total VFA and total protein production, but no significant difference ($P > 0.05$) on NH₃ production. Total VFA production of T0, T1, T2 and T3 treatments were 80.00; 112.50; 105.00 and 107.50 mM, respectively. NH₃ production of T0, T1, T2 and T3 treatments were 5.35; 6.29; 5.76 and 6.47 mM, respectively. Total protein production of T0, T1, T2 and T3 were 403.70; 471.44; 419.76 and 464.00 mg/g, respectively. It was concluded that 5% buffalo rumen content supplementation in complete feed resulting the best feed fermentability.

1. Introduction

Sheep is one of ruminant livestock raised by many people in Indonesia as a meat and wool producer. The advantages of sheep as livestock are small body size, easy to maintain, quickly reach sexual maturity, can give birth 3 times in 2 years, more than one litter size and good environmental adaptation [1]. One effort to increase livestock productivity is to provide feed that is suitable to livestock requirement.

Rumen content contains cellulolytic bacteria and other bacteria such as lignolytic and hemicellulolytic that can digest fibers. Rumen content can reach 8 – 10% of ruminant total body weight. Rumen content is feed consumed by ruminant contained in rumen and its nutrients have not been completely digested. Besides, rumen content contains many rumen microbes. Buffalo rumen content contains 102 – 180 CFU/g fungal colonies [2]; 10^{10} - 10^{11} cells/ml [3] total bacteria and total cellulolytic bacteria ranges from 3.74×10^7 - 10.9×10^7 /ml [4]. Probiotics were feed additives in the form of live microbes that can increase productivity and animal health. Requirements for bacteria can be regarded as probiotics i.e. able to survive in the digestive tract of livestock, do not harm to the host, and at least have 106 - 108 CFU/ml bacteria population [5]. The use of rumen microorganisms as probiotics can give a synergistic effect on the digestion of feed fiber [6].

The objective of the research is to examine the effect of different level of complete feed supplemented with buffalo rumen content as probiotic on the production of volatile fatty acids (VFA), ammonia (NH₃) and total protein production in Balibul sheep. The benefit of this research is to utilize buffalo rumen content as a slaughterhouse waste as a probiotic to increase Balibul sheep productivity.



2. Materials and Methods

The study was conducted from at Faculty of Animal and Agriculture Science, Diponegoro University, Semarang. Proximate analysis of feed ingredients and analysis of VFA, NH₃ and total protein were carried out at Nutrition and Feed Science Laboratory, Faculty of Animal and Agriculture Science, Diponegoro University, Semarang. Materials used in this study were 16 heads of male local sheep under 5 months of age (Balibul) with average body weight 11.58 ± 0.77 kg (CV = 6.61%). Equipments used during the study were individual cages, scales for weighing sheep and feed, buckets for feed and drinking water, chopper and vacuum pump for taking sheep's rumen fluid and a set equipment to analyze VFA, NH₃ and total protein production.

2.1. Preparation Stage

Preparation stage of research was carried out for 4 weeks including preparation of the cage, procurement of feed ingredients, procurement of rumen content, analysis of feed ingredients, formulation of complete feed, complete feed making, and procurement of 16 heads of Balibul male local sheep. Buffalo rumen content was obtained from Slaughterhouse at Kudus Regency, then dried forming air dry and grind using a grinder. Proximate analysis of feed ingredients was carried out at Nutrition and Feed Science Laboratory, Faculty of Animal and Agriculture Science, Diponegoro University, Semarang. Rations were prepared isoenergy and isoprotein with a protein content of 14% and TDN 66%. The formulation and nutrient content of complete feed can be seen in Table 1.

2.2. Adaptation

Balibul local sheeps were adapted to the condition of environment of the cage and treatment feed given. Adaptation phase was carried out for 4 weeks by providing treatment feed given little by little every day. Sheeps were injected with anthelmintic, antiparasitic and B-complex vitamins.

Table 1. Formulation and Nutrient Content of Complete

Feed Ingredient	T0	T1	T2	T3
	------(%)-----			
Cassava waste	7	7	5	3
Rice bran	20	17	8	8
Milled corn	4	4	4	5
Pollard	9	9	14	12
Molasses	3	3	3	3
Skim Milk	3	3	3	8
Mineral	2	2	2	2
Rumen content	0	5	10	15
Soybean meal	19	19	16	22
Coconut meal	8	11	15	6
Napier grass	25	20	20	16
Total	100	100	100	100
Nutrient Content (%)				
Dry matter	70.47	74.38	74.48	77.68
Ash	8.65	9.70	10.11	11.44
Crude protein	14.09	14.01	14.05	14.08
Crude fiber	28.65	28.84	30.14	27.86
Ether extract	6.02	5.71	4.73	4.90
Nitrogen-free extract	42.59	41.74	40.97	41.72
Total digestible nutrients	67.10	66.95	66.56	66.06

2.3. Preliminary Stage

Preliminary phase was carried out for 2 weeks, namely by giving feed treatment as much as 5% of body weight in order to find out how much feed can be consumed by livestock. Sheep were located in cage randomly according to treatment.

2.4. Treatment Stage

Treatment phase was carried out for 2 weeks, namely by providing treatment feed and drinking water ad libitum, then recording feed consumption. Livestock weighing was done once a week to find out average daily gain.

2.5. Data Collection Stage

Data collection phase was carried out for 1 day on the last day of treatment by taking rumen fluid. Rumen fluid was taken after 4 hours of feeding using a vacuum pump and then put in a bottle and stored in a fridge. Rumen fluid was analyzed for VFA, NH₃ and total protein at Nutrition and Feed Science Laboratory, Faculty of Animal and Agriculture Science, Diponegoro University, Semarang. VFA was analyzed by steam distillation method. NH₃ was analyzed using Conway microdiffusion technique and total protein was analyzed according to Kjeldahl method. VFA, NH₃ and total protein production were calculated using formula:

$$\text{VFA total (mM)} = (\text{ml titrant of blank} - \text{ml titrant of sample}) \times \text{N HCl} \times 1000/5$$

$$\text{NH}_3 \text{ total (mM)} = (\text{ml titrant of sample} \times \text{N H}_2\text{SO}_4 \times 1000)$$

$$\text{Total Protein (mg/g)} = \{(\text{ml titrant of HCl} - \text{ml titrant of blank}) \times \text{N HCl} \times 14 \times 6,25\} / \text{sample weight}$$

2.6. Experimental Design

Experimental design used in this study was a completely randomized design (CRD) consisting of 4 treatments with 4 replications. Treatments used were administration of different level buffalo rumen contents as probiotic, i.e. T0 (complete feed without buffalo rumen content), T1 (complete feed + 5% buffalo rumen content), T2 (complete feed + 10% buffalo rumen content) and T3 (complete feed + 15% buffalo rumen content). Variables observed were total VFA, NH₃ and total protein production. Collected data were analyzed using analysis of variance then proceed with Duncan's multiple range tests [7].

3. Results and Discussion

Results of the research concerning the effect of giving different level of buffalo rumen content on fermentability of Balibul local sheep presented in Table 2.

Table 2. VFA, NH₃ dan Total Protein Production

Variables	Treatments			
	T0	T1	T2	T3
VFA (mM)	80.00 ^b	112.50 ^a	105.00 ^a	107.50 ^a
NH ₃ (mM)	5.35	6.29	5.76	6.47
Total Protein (mg/g)	403.70 ^b	471.44 ^a	419.76 ^{ab}	464.00 ^a

Different superscripts on the same row indicate significant differences (P<0.05)

3.1. Volatile Fatty Acids (VFA) Production

Result of variance analysis showed that treatment of rumen contents supplemented with different level significantly affected (P <0.05) total VFA production of Balibul sheep. The average of total VFA production of each treatment were 80.00 mM (T0), 112.50 mM (T1), 105.00 mM (T2) and 107.50 mM (T3). Result of Duncan's multiple range test on mean value of treatment showed that T0 treatment was significantly different (P <0.05) with total VFA production in T1, T2 and T3 treatments, but total VFA production between T1, T2 and T3 treatments were not significantly different. The highest total VFA production was found in T1 treatment (112.50 mM) by giving 5% rumen content and the lowest total VFA production was found in the T0 treatment (80 mM). T0 treatment produced the lowest total VFA

production because there was no addition of buffalo rumen content as probiotic so that the ability to degrade energy source was low. Increasing of VFA can be influenced by the administration of probiotics that can help the process of degradation of organic compounds [8].

T1 treatment was not significantly different with T2 and T3, and only has a numerical decrease in value which indicated that increasing the amount of probiotic in rumen content in feed could not increase VFA production of sheep. VFA production in T1, T2 and T3 treatments were not significantly different because TDN content of feed was relatively the same so that an increase in the amount of buffalo rumen content as probiotic could not increase VFA production. TDN content is an overview of the amount of energy contained in digestible feed. TDN content of ration which relatively the same resulted almost the same of VFA production [9].

3.2. Ammonia Production (NH_3)

Results of analysis of variance showed that supplementation of different level of buffalo rumen content had no significant effect ($P > 0.05$) on the ammonia production (NH_3) of balibul sheep. Average of NH_3 production of treatments were 5.35 mM (T0), 6.29 mM (T1), 5.76 mM (T2) and 6.47 mM (T3), respectively. Probiotic did not affect NH_3 production because protein content and ration composition for each treatment were relatively the same, so that the protein degraded by rumen microbes to ammonia was relatively the same. The apparent rate of NH_3 production by mixed ruminal microorganisms depended on the substrate, the concentration of substrate, and the method used [10]. Factors that influence NH_3 production are feed protein content and protein degradability [11]. Ammonia can be originated from the breakdown of feed protein and from non nitrogen protein.

The average of ammonia production for each treatment has a range between 5.35 - 6.47 mM, these results were relatively the same as [12], which ranged from 5.26 to 6.64 mM in sheep fed with the addition of rumen microbial probiotics and catalytic supplements. Optimal ammonia concentration to support rumen microbial protein synthesis ranges from 4 - 12 mM [13].

3.3. Total Protein Production

Result of analysis of variance showed that supplementation of buffalo rumen content with different levels had a significant effect ($P < 0.05$) on the total protein production of Balibul sheep. The average of total protein production for each treatment was 403.70 mg/g (T0), 471.44 mg/g (T1), 419.76 mg/g (T2) and 464.00 mg/g (T3). Result of Duncan's multiple range tests on the mean of treatment showed that total protein production of T0 treatment was significantly different T1 and T3 treatments, but was not significantly different with T2 treatment. Total protein production of T1 treatment was significantly with T0 treatment, but not significantly different with the T2 and T3 treatments. Total protein production of T2 treatment was not significantly different with T0, T1 and T3 treatments.

The highest total protein production was found in T1 treatment with the provision of buffalo rumen contents by 5%, namely 471.44 mg/g and the lowest production in T0 treatment, namely 403.70 mg/g. Low of total protein production in T0 treatment was due to the absence of supplementation of buffalo rumen content as probiotic so that microbial protein produced in the rumen was low. Probiotic in feed will cause the number of microbes in the rumen to increase [14]. Therefore, addition of buffalo rumen content as probiotic will increase population of rumen microbes which in turn can increase fermentation of carbohydrates into VFA. VFA is used as a source of carbon skeleton for the formation of rumen microbial body. Rumen microbes derive most of their energy from the fermentation of carbohydrates in the form of VFA [15]. Rumen microbes that ferment pectin, starch and sugar use ammonia, peptides and amino acids for protein synthesis [16]. Thus, addition of probiotics can increase total protein. Besides, feed in the form of a mixture of forage and concentrate will increase microbial protein synthesis, due to increased synchronization of nutrient release, a better ruminal environment for rumen bacterial species diversity, increased number and type of substrate, increased feed consumption rates, and increased solid and liquid part diversity [17].

Total protein is obtained from undegradable feed protein and microbial protein, the more the number of microbes in the rumen, the more total protein. The formation of microbial protein will run optimally

if the production of ammonia, a high source of carbon and energy occur at the same time [18]. High of total protein production will be used by livestock for their living needs and for production. Total protein is a description of the availability of protein that can be used by ruminants which is influenced by protein content of feed, non-degraded feed, ammonia, carbon bonds and energy in the form of ATP in the rumen.

4. Conclusion

Based on the results of the study, it was concluded that supplementation of buffalo rumen content as probiotics in complete feed could increase feed fermentability, shown on the increasing of VFA, NH₃ and total protein production. Supplementation of 5% buffalo rumen content as probiotic was the best probiotic level.

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