Rumen Fermentation Characteristics and Methane Production in Sheep Fed a Total Mixed Ration Containing Coffee Residue

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Abstract

The objectives of this experiment were to determine the effect of coffee residue (CR) as a replacement of kleingrass hay in total mixed ration (TMR) on rumen fermentation characteristics and methane production in sheep. Four wethers with initial body weight (46 ± 7.2 kg) were used in a 4 × 4 Latin square design. The wethers were allotted to one of four TMR at 55 g dry matter (DM)/kg body weight -0.75/day. Four TMR used in this experiment were (CTL) Kleingrass hay + wheat + oat (50 : 25 : 25, on DM basis) as control; (LCR) : Kleingrass hay + wheat + oat + CR treated Bio-PKC (45 : 25 : 25 : 5, on DM basis); (MCR) Kleingrass hay + wheat + oat + CR treated with Bio-PKC (40 : 25 : 25 : 10, on DM basis); (HCR) Kleingrass hay + wheat + oat + CR treated with Bio-PKC (30 : 25 : 25 : 20, on DM basis). Bio-PKC was applied at level of 2% (w/w). The result showed that rumen pH value was maintained from 6.05 to 6.25. There was no significant (P>0.05) difference in rumen pH value by inclusion of coffee residue into TMR. The NH3-N concentration and protozoa number were similar (P>0.05) among treatments. There was no difference in methane production among treatments when expressed as l/kg DMI and l/day/BW-0.75. The study suggests that forage can be replaced by coffee residue treated with BIO-PKC up to 20% of the diet DM in ruminant feed.

Key words: total mixed ration, coffee residue, methane, sheep, rumen

Introduction

Appropriate use of relatively inexpensive agricultural and industrial by-products is of paramount important for profitable livestock production. However, high cost and low availability of conventional livestock feedstuff frequently demand consideration of by-product even if efficiency of utilization is low. In the beverage industry, wastes from coffee grounds have increased rapidly in recent years. Approximately 200.000

ton of coffee ground are produced annually in Japan. Although a small proportion of those wastes are converted into raw compost material, most are generally incinerated (Wakasawa *et al*., 1998; Xu *et al*., 2007).

There is increasing demand for the efficient use of food by-products because of economic and environmental concerns. Coffee grounds usually contain 14.5% CP, 18.4% ether extract (EE), 68.8% neutral detergent fiber (NDF), and 54.8% acid detergent fiber (ADF) (Xu *et al*., 2007). Santoso *et al*. (2011) reported that coffee waste contain 13.2 of crude protein (CP), 68.1% of NDF, 45.2% of ADF, 16.5% of EE and 0.08% of caffeine. Therefore, coffee ground could possibly be a source of nutrients for ruminants Xu *et al.* (2007).

It is difficult to recycle those wastes as animal feeds, because they contain high moisture and considerable amount of secondary metabolite such as caffeine and tannin which may reduce appetite and protein digestibility of the feeds. However, the secondary metabolites have been reported to mitigate rumen methane emission. In the previous study, Santoso *et al*. (2011) reported that *in vitro* CH_4 production in coffee waste substrate was lower by 70% as compared timothy grass hay substrate. Therefore, the fermented residues processed to prevent aerobic deterioration with the mixed microbial products might be significant means in environmental and resource recycling aspects for sustainable agriculture. The objectives of this experiment were to determine the effect of coffee residue treated by Bio-PKC as a replacement of kleingrass hay in TMR on rumen fermentation characteristics and methane production in sheep.

Materials and Methods

Animal and Treatments

Four wethers with initial body weight (46 ± 7.2 kg) were used in a 4 × 4 Latin square design. The wethers were individually housed in metabolic cages and fed the four total mixed ration (TMR) at 55 g DM/kg body weight $(BW)^{-0.75}$/day to meet maintenance energy requirements. Half of the TMR were fed at 08:00 h and the other half at 16:00 h. Water and sodium chloride block were freely available throughout experiment. Four TMR were used in this experiment, namely: CTL : Kleingrass hay + wheat + oat (50 : 25 : 25, on DM basis) as control; LCR : Kleingrass hay + wheat + oat + CR treated with Bio-PKC (45 : 25 : 25 : 5, on DM basis); MCR: Kleingrass hay + wheat + oat + CR treated with Bio-PKC (40 : 25 : 25 : 10, on DM basis); HCR: Kleingrass hay + wheat + oat + CR treated with Bio-PKC (30 : 25 : 25 : 20, on DM basis). The BIO-PKC was applied at level of 2% (w/w). The experiment was conducted in 4 periods with four wethers per treatment per period. Each period was consisted of 7 days adaptation, 2 days respiratory trial and followed by 1 day for rumen fluid collection. Refusals were weighed daily before the afternoon feeding. Body weight was measured before the afternoon feeding at the beginning and end of each period.

Respiratory Trial

Respiratory trial is conducted during 2 days of each period. Oxygen consumption, and carbon dioxide and methane production by each animal are monitored by an open circuit respiratory system using a hood over the animal's head.

Rumen fluid collection

Rumen fluid (20 ml) were collected from each wether via fistula by using a 50 ml hand syringe immediately before feeding (0) and at 1, 2, 4, 6, 8 h after feeding on the last day of each period. The pH and ORP was measured immediately by using a pH meter (D-51, Horiba Ltd., Japan). Sample was frozen at –10 °C for further analysis of ammonia nitrogen and volatile fatty acids (VFAs) concentrations. Concentrations of individual VFAs were analyzed using a gas chromatography (GC 2014, Shimadzu, Japan). Concentration of NH_3-N was analyzed according to method of Conway and O'Malley (1942). One millilitre of rumen fluid was mixed with 4 ml of Methyl Green Formalin Saline (MFS) to count protozoa number.

Sample Analyses

Dried samples were used to determine DM, ash and CP according to procedure of AOAC (1995). Procedure of Van Soest *et al.* (1991) was used to determined concentrations of NDF, ADF and acid detergent lignin (ADL). NDF was determined without the use of ∝-amylase and sodium sulphite.

Statistical Analysis

Data of gas emission and fermentation characteristics were subjected to analysis of variance for a Latin square design using GLM procedure of SAS (SAS Institute Inc., Cary, NC). When significant effects (*i.e.*, $P<0.05$) of the treatment occurred, Duncan's multiple range test were used to determine differences between treatments. Significance was declared at $P<0.05$, and a tendency toward significance was declared at $0.05<P<0.10$.

Results and Discussion

The chemical composition of TMR used in this experiment is shown in Table 1. Increasing concentration of coffee residue in the TMR tended to reduce DM, hemicellulose contents and to increase ADF content. The OM, CP, cellulose and GE contents of all TMR were similar 94.3 to 95.5%, 13.8 to 13.9%, 14.9 to 15.0% and 19.1 to 20.0%, respectively. In a previous study, Santoso *et al*. (2011) reported that coffee waste contained 0.08% of caffeine. Similar value of 0.13% of caffeine in coffee grounds has been reported by Bartley *et al*. (1978).

Table 2 shows methane production in sheep fed TMR containing coffee residue. There was no significant ($P>0.05$) difference in methane production among

Table 1. Chemical composition of experimental TMR containing coffee residue

	TMR			
	CTL	LCR	MCR	HCR
DM (%)	88.3	83.9	79.6	70.9
	----------% of DM----------			
OM	94.3	94.6	94.9	95.5
CP	13.9	13.9	13.8	13.8
NDF	47.0	47.1	47.3	47.7
ADF	17.5	18.5	19.5	21.5
Hemicellulose	29.5	28.7	27.8	26.1
Cellulose	14.9	14.9	14.9	15.0
GE (MJ/kg of DM)	19.1	19.3	19.5	20.0

Table 2. Methane production in sheep fed TMR containing coffee residue

	TMR				SEM	P
	CTL	LCR	MCR	HCR		
CH_4 (l/d/kg $BW^{0.75}$)	1.49	1.36	1.55	1.63	1.44	0.79
CH_4 (l/kg DMI)	26.61	28.77	29.45	30.32	0.44	0.70

treatments when expressed as l/kg DMI and l/day/$BW^{0.75}$. Increasing concentration of coffee residue in TMR resulted in a higher methane production when expressed as l/day/$BW^{0.75}$. This result could be attributed to increased ADF content in the TMR. Moss (1994) revealed that digestible ADF, cellulose and hemicellulose are important variables influencing CH_4 production in the rumen. In a previous study, Santoso *et al*. (2011) found that *in vitro* CH_4 production in coffee waste substrate was lower by 70% as compared timothy grass hay substrate.

Table 3 summarizes the rumen pH value, concentrations of NH_3-N and VFA in the rumen of sheep fed the TMR containing coffee residue. Inclusion of coffee residue in the TMR had no effect (P>0.05) on pH value, protozoa number, concentrations of NH_3-N, total VFA, acetate and propionate. Average of pH values in the rumen of sheep fed coffee residue varied from 6.06 to 6.26, which are in the optimal pH range of 6.7 ± 0.5 required to maintain normal cellulolysis (Van Soest, 1994) and required for microbial protein synthesis (Russell *et al*. 1992). Total VFA concentration in the rumen of sheep fed TMR containing coffee residue (LCR, MCR and HCR) was relatively higher than of the control sheep. This result suggesting that inclusion coffee residue up to 20% of DM in TMR did not inhibit rumen microbial fermentation. This finding was also supported by there was no significant

Table 3. Rumen fermentation characteristics in sheep fed TMR containing coffee residue

	TMR				SEM	P
	CTL	LCR	MCR	HCR		
pH	6.20	6.26	6.06	6.17	0.05	0.18
N-NH_3 (mg/100 ml)	74.9	75.8	73.1	69.7	20.48	0.99
Protozoa (10^6 cell/ml)	1.5	1.1	1.2	1.6	0.17	0.23
Total VFA (mM)	60.7	75.0	68.0	78.7	7.38	0.40
Acetate (mol/100 mol)	78.0	73.1	78.4	75.2	1.95	0.28
Propionate (mol/100 mol)	18.1	23.7	18.0	19.4	1.66	0.15
Butyrate (mol/100 mol)	2.7[a]	1.9[b]	2.4[a]	2.7[a]	0.15	0.02

Means in the same row followed by different letters are different (P<0.05)

effect on DM digestibility of sheep due to inclusion coffee residue in TMR (data not shown). In the previous study, however, Bartley *et al*. (1978); Xu *et al*. (2007) reported that the ruminal fluid from Holstein steers or sheep receiving coffee ground had significantly lower concentration of total VFA than those receiving no coffee ground. Proportion of butyrate in sheep fed LMR was (P<0.05) lower than those fed other TMR.

Conclusion

Rumen methane production was similar in sheep fed all TMR. Total VFA was relatively higher in sheep fed TMR containing coffee residue than those fed TMR without coffee residue. Forage can be replaced by coffee residue treated with Bio-PKC up to 20% of the diet DM in ruminant feed.

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