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Digestibility, Fermentation Characteristic, Protein Microbial Synthesis and Growth Performance of Beef Cattle Fed High Forage Ration with Lerak Extract Supplementation

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Abstract: This research was aimed to investigate the utilization of whole lerak extract to improve rumen fermentation, nitrogen retention and performance of beef cattle received high forage based ration. Experimental diet composed of forage (70%) and concentrate (30%). The *in vivo* study was conducted using 12 local beef cattle which were divided into three treatments ie three different levels of lerak extract (0, 100 and 200 mg/kg body weight) were added to the diet. Parameters measured were nutrient digestibility, volatile fatty acid (VFA) profile, NH_3 concentration, microbial protein synthesis, feed intake and daily gain of beef cattle during 90 days of feeding trial. The addition of lerak extract up to the level of 200 mg/kg BW did not affect nutrient digestibility. Total VFA and propionate proportion increased ($p<0.05$) and the ratio of acetate: propionate decreased ($p<0.05$) with the addition of lerak extract. Concentration of NH_3 in the rumen tended to decrease. Nitrogen retention, microbial protein synthesis, feed intake and daily gain of local beef cattle fed high forage ration tended to increase with the addition of lerak extract at the level up to 200 mg/kg BW. The addition of lerak extract at the level of 200 mg/kg BW increased average daily gain up to 12.5% compared to the control treatment.

Key words: Digestibility, daily gain, rumen fermentation, *Sapindus rarak*, ongole beef cattle

INTRODUCTION

Production of local beef cattle in Indonesia is still low because the farmers fed the cattle with high forage-based rations. This condition is different from feedlot system that fed their cattle with high concentrate feed (70-90% dry matter). The high use of low quality forage caused deficiency of nutrients, especially protein/ nitrogen. Defaunation treatment using saponin is one of the strategies to overcome nitrogen deficiency as saponin can suppress the growth of protozoa, increase microbial protein supply for cattle and modify rumen fermentation.

Lerak extract (*Sapindus rarak*) that contains high saponin (81.47%) reduced the growth of protozoa *in vitro* and *in vivo* (Wina *et al*., 2006; Suharti *et al*., 2011). Previous study using *in vitro* fermentation in the different ratios of forage and concentrate showed that saponin lerak extract at the level of 0.8 mg/ml markedly reduced protozoan numbers in all diets, slightly increased *Ruminococcus albus* number, significantly enhanced *Prevotella ruminicola* number and increased propionate concentration in all diets after 24 h incubation (Suharti *et al*., 2011).

It has been known that propionate is a major energy source in ruminant especially beef cattle. The increased propionate synthesis with the addition of lerak extract has a potency to be used to improve the performance of beef cattle fed with high forage ration. The results of *in vitro* studies need to be followed by *in vivo* study to evaluate the effect of the use of lerak extract on the performance of local beef cattle fed high forage ration.

This present study was designed to evaluate the use of lerak on the digestibility, fermentation characteristics, protein microbial synthesis and performance of beef cattle fed high forage ration (70%).

MATERIALS AND METHODS

Cattle, experimental design and treatments: The experiment was carried out at the Field Laboratory, Department of Nutrition and Feed Technology, Faculty of Animal Science, Bogor Agricultural University, Indonesia. A total of 12 male Ongole Crossbreed beef cattle with initial body weight (BW) of 187.5±13 kg, were assigned into three dietary groups which each consisted of four animals.

The cattle were housed in individual tie stall and had free access to water during the experiment. The experimental conditions and animal procedures were handled in accordance with guidelines described by the Meat and Drought Animal Nutrition Laboratory at Faculty of Animal Science, Bogor Agriculture University. All groups received the same basal diet consisting of native

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Table 1: Nutrient composition of native grass, concentrate mix and total mix ration for *in vivo* feeding (% dry matter)

Nutrient	Native grass (G)	Concentrate Mix (C)	Total mix ration (G:C = 70:30)
Ash	11.11	13.09	11.70
Crude protein (CP)	10.20	20.05	13.16
Crude fiber (CF)	40.12	21.42	34.51
Ether extract (EE)	0.45	3.14	1.26
BETN	38.12	42.50	39.43
Calcium	0.38	1.88	0.83
Phosphorus	0.14	0.84	0.35
TDN	46.64	65.38	51.96

TDN: Total digestible nutrient = 92.64 - 3.338 (CF) - 0.945 (EE) - 0.762 (BETN) + 1.115 (CP) + 0.031 $(CF)^2$ - 0.133 $(EE)^2$ + 0.036 (CF) (BETN)+ 0.207 (EE)(BETN)+0.100(EE)(CP)-0.022$(EE)^2$(CP) (Hartadi *et al*., 1980)

grass and concentrate mix with the ratio of 70:30. Concentrate mix (self mixed) consisted of soybean meal (6%), coconut cake meal (30%), cassava waste (18.5%), wheat pollard (35%), molasses (5%), $CaCO_3$ (3%), DCP (0.5%), NaCl (0.5%), premix (0.5%) and urea (1%). The premix composition consist of Vitamin A 500000 IU, Vitamin D 100000 IU, Vitamin E 150 mg, Vitamin B1 50 mg, Vitamin B2 250 mg, Vitamin B12 250, mcg, Vitamin K 50 mg, Niacinamide 375 mg, Ca-d-Panthotenate 125 mg, Folic Acid 25 mg, Choline Chloride 5000 mg, L-lysine 3750 mg, Dl-Methionine 5000 mg, Mg Sulfate 1700 mg, Fe Sulfate 1250 mg, Mn Sulfate 2500 mg, Cu Sulfate 25 mg, Zn Sulfate 500 mg, K Iodine 5 mg, Antioxidant and Carrier per 1 kg. The nutrient composition of the ration was shown in Table 1.

The experiment was designed as a 3 x 4 block randomized design with 3 dietary treatments such as: (1) basal diet+0 mg lerak extract (control), (2) basal diet+lerak extract with dosage of 100 mg/kg BW and (3) basal diet+lerak extract with dosage of 200 mg/kg BW. The lerak extracts were administered as a meal, prepared by mixing with concentrate mix. The cattle were housed in individual tie stall. All animals received approximately 2.8-2.9% of body weight of total mixed ration (TMR) and allowed to the experimental feeding for 90 days. The diets were offered *ad libitum* and were fed twice daily. Samples of the total mixed ration and feed remnant were collected for proximate, NDF and ADF analysis.

Sampling and measurement: Source of rumen fluid was taken by using stomach tube at 4 h after feeding on 30, 60 and 90 days after treatment. Rumen fluid was filtered by using cheese cloth and the supernatant obtained was kept for protozoa counting, VFA profile and ammonia analysis.

Total VFA concentrations and molar proportions of VFA were analyzed by using gas chromatography (Chrompack CP9002, Netherlands, flame ionized detector, Capillary column type WCOT Fused Silica 25 m x 0.32 mm, oven temperature: conditioning at 60°C and running at 115°C and nitrogen as gas carrier). Before analysis, the pH of the rumen fluid aliquots from *in vitro* incubation was adjusted to 3-4 with H_2SO_4. Thus, 1.5 ml of the adjusted rumen fluid was mixed with 30 mg sulfosalicylic acid ($C_7H_6O_6S.2H_2O$) and centrifuged at 12,000 rpm x 10 min (7°C) then 0.5 µl of the mixed solution was injected to the GC. Ammonia (N-NH_3) concentrations were determined by using the micro diffusion method (Conway, 1962). The numbers of protozoa in the rumen fluid were counted under a microscope. 0.5 ml rumen fluid was mixed with 0.5 ml methyl green formaldehyde saline solution containing 35% formaldehyde, distilled water, methyl green and NaCl. The population of protozoa was counted directly by using a counting chamber (0.1 mm) under a microscope (40x).

Nutrient digestibility was measured by fecal total collection method during 5 consecutive days in the middle of the treatments. Fecal samples (10% w/w of wet weight) were composited from each cattle and subsequently dried at 60°C in the oven for dry matter (DM), organic matter (OM), crude fiber (CF), crude protein (CP) and ether extract (EE) analysis. The digestibility of DM was calculated as follows: DM digestibility (%) = ((DM Intake - fecal DM) / DM intake) x 100%. The digestibilities of OM, CF, CP and EE were calculated by using the same formula with the corresponding intake of each nutrient.

Nitrogen retention was measured by analyzing nitrogen concentration in the urine. Urine collection was done during 5 consecutive days concurrently with fecal collection. The urine sample was acidified with 250 ml/10 L of H_2SO_4 10% for analysis of nitrogen and allantoin concentration. Allantoin concentration was used to estimate total purine derivatives (PD) with approach that allantoin concentration = 82.5% of total PD (IAEA, 1997) and then total PD (mmol/d) was calculated by 100/82.5 x allantoin concentration.

The total PD was used to estimate purine absorption of Ongole beef cattle (Makkar and Chen, 2004) by using formula as follows: Y = 0.85X + $(0.132BB^{0.75})$, where 0.085 = proportion of PD in the plasma and excreted through urine, 0.132 = purine endogen derivative which excreted through urine (mmol/kg $BB^{0.75}$), Y = excreted PD (mmol/day), X = purine absorption (mmol/day).

Purine absorption was used to estimate microbial protein synthesis by using approach as follows: N microbe (g/day) = 70X/(0.116x0.83x1,000) = 0.727 X, where 70 = N purine (mg/mmol), 0.83 = digestibility coefficient of N microbe, 0.116 = N purine: total N ratio of microbe biomass of Ongole beef cattle and X = absorption of PD in mmol/d. Growth performance of Ongole beef cattle was analyzed by measuring daily feed intake, body weight gain and feed efficiency.

Data analysis: Statistical analysis of the data was carried out by ANOVA using General Linear Procedure. If there were any different among treatments further tested by using Duncan's multiple range test was conducted. Computation was performed by using SPSS 13.0 for windows evaluation version.

RESULTS AND DISCUSSION

Effect of lerak extract on rumen protozoa: The addition of lerak extract up to 200 mg/kg BW in the ration of Ongole Crossbreed beef cattle did not significantly affect the population of rumen protozoa during 90 days treatment (Table 2). Thirty days after treatment, the addition of lerak extract at the level of 100 mg/kg BW had a slight reduction of protozoa population i.e., 12%, while the addition of lerak extract at the level of 200 mg/kg BW reduced protozoa population up to 15%. At the 60 days after treatment, the addition of lerak extract at the level of 100 mg/kg BW and 200 mg/kg BW reduced protozoa population by 19 and 37%, respectively and similar reductions were observed until 90 days after treatment. These results were contrary with the result of previous *in vitro* study that showed the significant reduction of protozoa population with lerak extract addition (Suharti *et al*., 2011). The different effects of lerak extract on protozoa population in the *in vitro* and *in vivo* studies might be due to the rumen flow and saponin adaptation by rumen ecosystem of host animal and will decrease the inhibition activity on the protozoa growth. Odenyo *et al*. (1997) reported that saponin of *Sesbania sesban* could reduce protozoa population when it was administered directly to the rumen, but did not have antiprotozoal effect when it was fed orally to the sheep. The adaptation of rumen microbe to saponin was one of the factors which caused wide variety of antiprotozoal activity of saponin (Wallace *et al*., 2002). However, Quillaja saponin is relatively stable in the rumen during 6 hours and has antiprotozoal activity (Makkar and Becker, 1997). The different results were reported by Abreu *et al*. (2004) that protozoa population was increased in sheep receiving *Sapindus saponaria* saponin. These results showed that rumen microbe can degrade saponin in the rumen into its aglycone and glycoside compounds. The saponin degradation process may be faster in the *in vivo* than in the *in vitro* studies therefore result in the different effect of saponin on protozoa population. Saponin degradation in the rumen has been reported by Wina (2005) who showed that degradation product of *S. rarak* saponin (aglycone structures) appeared at 2 h after feeding and hederagenin as aglycone at 4 h after feeding.

Effect of lerak extract on nutrient digestibility and fermentation characteristics: The addition of lerak extract up to the level of 200 mg/kg BB in the ration of Crossbred Ongole beef cattle fed high forage, did not significantly affect nutrient digestibility such as DM, OM, CP, CF and EE digestibilities compared to the control treatment (Table 3). In contrast, total VFA production and propionate proportion were significantly increased ($p<0.05$) and acetate:propionate ratio was significantly decreased ($p<0.05$).

Nutrient digestibility (DM, OM, CP, CF and EE) was not affected by the addition of lerak extract indicating that saponin in the lerak extract did not alter digestion activity of rumen bacteria. The downturn of protozoa population due to lerak extract addition, did not affect nutrient digestibility. The different results have been reported by Abreu *et al*. (2004) that intraruminal supplementation of *Sapindus saponaria* (12% saponin) at the level of 8 g/kg $BB^{0.75}$ did not affect on organic matter and protein digestibility, however ADF digestibility decreased by 10% and acetate:propionate ratio was also decreased.

The reduction of NH_3 production with the addition of lerak extract at the level of 200 mg/kg BW was related to the defaunation activity of saponin contained in the lerak extract. Protozoa are proteolytics, therefore the inhibition of protozoa activity will decrease feed protein degradation and also ammonia production. The rate of feed protein degradation and non-protein nitrogen also determine the ammonia concentration in the rumen. In addition, the inhibition of protozoa might increase the use of ammonia by rumen bacteria for protein bacteria

Table 2: Protozoa population of rumen ongole beef cattle supplemented lerak extract in the ration during 90 days treatment

	---------- Lerak extract level (mg/kg BW) ----------			
Days after treatment	0	100	200	SEM
Protozoa population (x 10^4/ml)				
30 d	9.7	8.5	8.2	1.22
60 d	6.8	5.5	4.3	1.53
90 d	7.0	5.1	4.3	0.74

Table 3: Nutrient digestibility and fermentation characteristics of ongole beef cattle supplemented lerak extract in the ration during 90 days treatment

	---- Lerak extract levels (mg/kg BW) ----			
Parameters	0	100	200	SEM
Nutrient digestibility (%)				
Dry Matter (DM)	58.2	53.5	54.9	1.88
Organic Matter (OM)	62.9	59.4	59.7	1.05
Crude Protein (CP)	73.4	71.4	71.1	0.84
Crude Fiber (CF)	75.9	74.0	74.6	1.08
Ether Extract (EE)	44.1	53.6	40.1	3.99
N-NH3 (mM)	7.47^{ab}	8.09^{a}	4.28^{b}	0.80
Total VFA(mM)	84.87^{b}	123.54^{a}	111.57^{a}	5.87
Proportion of VFA(% total VFA)				
Acetate	66.77	64.88	65.14	0.46
Propionate	16.83^{b}	18.83^{a}	19.07^{a}	0.35
Iso-butyrate	2.29	2.38	2.58	0.13
Butyrate	11.92	11.45	10.87	0.29
Iso-valerate	1.53	1.48	1.38	0.06
Valerate	0.67	0.98	0.96	0.09
Acetate:propionate ratio (A:P)	3.97^{b}	3.45^{a}	3.41^{a}	0.09

Different superscripts on the same row represents a significant difference ($p<0.05$)

Table 4: Estimation of microbial protein synthesis (MPS) of Ongole beef cattle supplemented with lerak extract in the ration

	Level of lerak extract (mg/kg BW)			
Parameter	0	100	200	SEM
Allantoin (mmol/d)	21.96	23.33	29.14	2.35
Purine derivative/DP (mmol/d)*	26.61	28.27	35.33	2.85
Purine absorption (mmol/d)	23.14	25.00	33.19	3.37
Microbial N supply (g/d)	16.82	18.18	24.13	2.45
Microbial protein synthesis/MPS (g/d)	105.15	113.61	150.81	15.30
MPS efficiency (g/kg FOM)	64.36	86.81	96.71	9.94

*Urine allantoin = 82.5% total PD (IAEA, 1997), FOM=Fermented organic matter in the rumen = 0.65x Degraded organic matter (IAEA, 1997)

synthesis and decreased rumen ammonia concentration. Supplementation of the methanol extract of the lerak fruit pericarp at the levels of 0.42 and 0.72 g/kg BW in the sheep rations composed of elephant grass and pollard (65:35) significantly reduced the NH_3 concentration (Wina *et al*., 2006). Hu *et al*. (2006) also reported that inclusion of tea saponins decreased the ammonia-N concentration by 8.3 and 19.6% in the faunated and defaunated rumen fluid, respectively. The addition of Quillaja saponin at concentration of 6 g/L also reduced ammonia concentration in the *in vitro* fermentation (Patra and Yu, 2014)

The increase in total VFA production and propionate proportion with the addition of lerak extract indicates an increase in efficiency of fermentation by rumen microbes. In addition, lerak extract can also modify rumen microbial activity by directing the formation of propionate and reduced butyrate production. This result is supported by previous *in vitro* data (Suharti *et al*., 2011) showing that lerak extract modified the composition of rumen bacteria which increased *Prevotella ruminicola*, the propionate and succinate-producing bacteria population in the rumen system. The increase in propionate production could reduce the supply of H_2 since the production of propionate in the metabolic pathway in the rumen uses H_2, which competes with methanogenic bacteria to form methane. Therefore, the use of lerak extract has a great potential to reduce methane production in the rumen. Previous study showed that tea saponin at level 5 g/kg DM significantly increased propionate, decreased acetate proportion and decreased methane emissions (Yuan, 2007). Across a range of forages and diets, the addition of Yucca schidigera extract at level 110 mg/kg diet which contain saponin consistently reduced methane production without influencing gas production kinetics or total VFA concentrations measured at 24 h *in vitro* fermentation (Xu *et al*., 2010).

Effect of lerak extract on microbial protein synthesis and nitrogen retention: The addition of lerak extract up to the level of 200 mg/kg BW did not affect ($p>0.05$) microbial protein synthesis in the rumen (Table 4). However, the efficiency of microbial protein synthesis in this study is still in the normal range i.e., 6.44-11.81 g/100 g fermented organic matter. Karsli and Russell (2001) reported that the efficiency of microbial protein synthesis in the rumen ranged between 7.0-27.9 g/100 g fermented organic matter depending on dry matter intake, forage and concentrate ratio, the rate of degradation of carbohydrates and N, synchronization of N and energy release and the rate of passage of feed. The low quality of forage will reduce the efficiency of microbial protein synthesis due to lower nutrient degradability and the availability of precursor for microbial protein synthesis.

Table 5: Nitrogen balance of ongole beef cattle supplemented with lerak extract in the ration

	-- Lerak extract levels (mg/kg BW) --			
Variable	0	100	200	SEM
N Intake(g/h/d)	97.07	92.10	97.13	1.93
N in feces (g/h/d)	25.80	26.13	28.01	0.58
Digestible N (g/h/d)	71.25	65.96	69.10	2.03
N urine (g/h/d)	23.67	19.75	18.25	1.24
N retention (g/h/d)	47.57	46.20	50.85	2.19
% N retention from N Intake	49.03	49.75	52.39	1.65
% N retention from digestible N	66.81	69.41	73.67	1.89

Table 6: Performance of ongole beef cattle supplemented with lerak extract in the ration during 90 days

	---- Level of lerak extract (mg/kg BW) ----			
Variables	0	100	200	SEM
Dry matter intake (kg/h/d)				
Forage (kg/h/d)	2.95	2.92	3.10	0.07
Concentrate (kg/h/d)	1.52	1.55	1.52	0.02
Total ration (kg/h/d)	4.47	4.46	4.62	0.08
ADG (kg/h/d)	480	500	540	16.56
Feed efficiency	0.11	0.12	0.13	0.005

ADG: Average daily gain

The addition of lerak extract up to 200 mg/kg BW did not significantly increase microbial protein synthesis in beef cattle fed high amounts of forage (native grass). Although the lerak extract increased VFA production which supplied the energy and carbon sources for precursor of the microbial protein synthesis, but the rumen NH_3 concentration was low (4 mM). This led to the imbalanced protein/energy (P/E) ratio, hence reduced microbial protein synthesis. In addition, the efficiency of microbial protein synthesis was also influenced by the concentration of trace minerals and vitamins (Karsli and Russel, 2001). In this study, the ration of beef cattle used high level of native grass which might be deficient in sulfur and phosphorus and might affect the efficiency of microbial protein synthesis. Mineral sulfur (S) has been known plays an important role in the growth of bacteria, especially for methionine and cysteine synthesis with levels ranging between 0.11-0.20% in the ration and depend on the animal health status (NRC, 1984). Mineral phosphor is required for the synthesis of ATP and protein by microbes (Stern and Hoover, 1979).

Some research reports showed that the effects of saponins on microbial protein synthesis vary considerably depending on the source of saponins and saponin levels used. The use of steroidal saponins (SAP) isolated from *Yucca schidigera* extract increased microbial protein synthesis at 15 μg/ml but decreased it by the higher concentrations in the *in vitro* fermentation (Wang *et al*., 2000). Wina *et al*. (2006) showed that the addition of S. rarak extract at level 0.72 g/kg body mass on sheep did not affect microbial N supply. In contrast, the addition of saponin from *Biophytum petersianum* Klotzsch at the level of 26 mg/kg BW of goats that received rations consisted of elephant grass and concentrate (70:30), could improve the efficiency of

microbial N synthesis up to 51% (Santoso *et al*., 2007). Mao *et al*. (2010) also reported the increase in microbial protein synthesis with the addition of tea saponin at level 3 g/d or tea saponin plus soy bean oil on growing lamb. The reduction of protozoa population could reduce bacterial protein degradation and will increase microbial protein flow to the intestine. It is known that protozoa have important role in microbial N cycle in the rumen (Jouany, 1996). The N flows in duodenum were affected by *Sapindus saponaria* (contain saponin) supplementation, except N flow from microbe (Abreu *et al*., 2004). In contrast, Goetsch and Owen (1985) stated that the addition of sarsaponin from *Yucca schidigera* at the level of 44 mg/kg on dairy cattle did not affect N microbial flow to the duodenum. Hristov *et al*. (1999) also reported that supplementation of *Y. schidigera* in meal fed to the heifers at the level of 20 and 60 g/day did not significantly affect N microbial.

The addition of lerak extract up to the level of 200 mg/kg BW did not significantly increase nitrogen retention (Table 5). The percentages of N retention from digestible N for all treatments were in the range of 66-73% indicating that the quality of feed protein was good and was utilized by Ongole beef cattle efficiently. The N retention from N intake on Ongole beef cattle which fed ammoniated rice straw was 65% (Hindratiningrum *et al*., 2009).

The trend of increasing microbial protein supply with supplementation of lerak extract in this study is in line with nitrogen retention. This suggests that there is a correlation between bacterial protein syntheses with N. The higher microbial protein supply will increase the N retention due to the high quality of microbial protein which have balanced amino acid composition.

Effect of lerak extract on performance of ongole beef cattle: The addition of lerak extract up to the level of 200 mg/kg BW did not significantly increase average daily gain (ADG) and feed efficiency of Ongole beef cattle fed high forage during 90 days treatment (Table 6). Although the addition of lerak extract already significantly increased total VFA production and the propionate proportion, the ADG was not significantly improved. As we know that volatile fatty acids have important role as a major glucose source in the ruminant since glucose from feed only provide less than 10% of glucose requirement (Yost *et al*., 1997). Furthermore, a major glucose source in the ruminant was gluconeogenesis pathway which used propionate as a major precursor and could provide approximately 27-59% carbon sources for host animal. In addition, amino acid, glycerol and lactate are also carbon sources (Cerilla and Martinez, 2003). Ratio of acetate to propionate in this study was 3.4:1 and could stimulate the body weight gain since optimal ratio of acetate: propionate to produce optimal body weight growth was 3:1 (Yost *et al*., 1977).

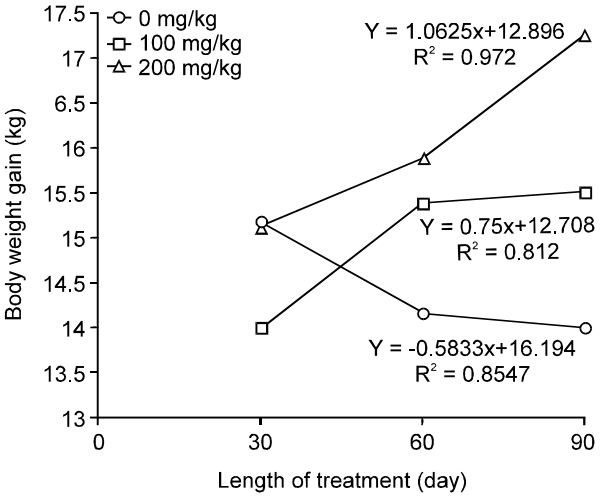

Fig. 1: Trend of body weight gain improvement of Ongole beef cattle supplemented with lerak extract in the ration during 90 Days)

However, with increasing the duration of treatment, there was improvement in body weight gain of Ongole beef cattle fed lerak extract at the levels of 100 and 200 mg/kg BW (Fig. 1). During 30 days of treatment, the addition of lerak extract did not increase the body weight gain of Ongole Beef Cattle. After 60 days and 90 days treatments, the addition of lerak extract tended to increase the body weight gain. During 90 days treatment, the addition of lerak extract at the levels of 100 and 200 mg/kg BW increased body weight gain of Ongol beef cattle up to 4.2 and 12.5%, respectively.

The trend of body weight gain improvement with time of feeding experiment indicating that the addition of lerak extract may have the beneficial effect as feed additive to stimulate the growth of beef cattle when fed for longer period.

The previous study showed the positive effect saponin supplementation on livestock performance. Navas-Camacho *et al*. (1993) reported that the addition of *E. cyclocarpum* leave meal (containing saponin) at the level of 100 g/d for sheep fed *Pennisetum clandestinum* could increase dry matter digestibility up to 17% and body weight up to 53% compared to the control treatment. However, the addition of *E. cyclocarpum* at high level (300 g/d) decreased dry matter digestibility. Thalib *et al*. (1996) also reported the improvement of sheep body weight up to 22% which received lerak extract per 3 days by using force feeding. Wina *et al*. (2006) suggest that the addition of lerak extract every day increased the ADG of sheep up to 40%. Tea saponin at the level of 3 g/d also could increased dry matter intake, ADG and feed efficiency of sheep compared to control treatment (Hu *et al*., 2006).

Conclusion: The addition of lerak extract (*Sapindus rarak*) at the level of 200 mg/kg BW to Ongole Cross-

breed cattle was beneficial for modifying rumen fermentation as it increased total VFA and propionate production and decrease acetate: propionate ratio as well as rumen ammonia. Addition of lerak extract at the level up to 200 mg/kg BW showed a linear increase of ADG of local beef cattle fed high forage ration with longer time of feeding experiment compared to the control treatment.

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REFERENCES

Abreu, A., J.E. Carulla, C.E. Lascano, T.E. Diaz, M. Kreuzer and H.D. Hess, 2004. Effects of *Sapindus saponaria* fruits on ruminal fermentation and duodenal nitrogen flow of sheep fed a tropical grass diet with and without legume. J. Anim. Sci., 82: 1392-1400.

Cerilla, M.E.O. and G.M. Martinez, 2003. Starch digestion and glucose metabolism in the ruminant: a review. Interciencia, 28: 380-387.

Cheeke, P.R., 2000. Actual and potential applications of *Yucca schidigera* and Quillaja saponaria saponns in human and animal nutrition. Proceed. of the Am. Soc. Anim. Sci., pp: 10.

Conway, E.J., 1962 Ammonia. General method. Microdiffusion Analysis and Volumetric Error. Crosby Lockwood and Son Ltd., London, UK., Page 98-100.

IAEA (International Atomic Energy Agency), 1997. Estimation of Rumen Microbial Protein Production from Purine Derivatives in Urine. Vienna Austria.

Goetsch, A.L. and F.N. Owens, 1985. Effects of sarsaponin on digestion and passage rates in cattle fed medium to low concentrate. J. Dairy Sci. 68: 2377-2384.

Hartadi, H., S. Reksohadiprodjo, S. Lebdosukojo, A. Tillman, L.C. Kearl and L.E. Harris, 1980. Tables of Indonesian feedstuffs composition. International Feedstuffs Institute Utah Agricultural Experiment Station, Utah.

Hindratiningrum, N., M. Bata and Suparwi, 2009. Microbial protein production and nitrogen balance of male local beef cattle fed amoniated rice straw. Anim. Prod., 11: 116-121.

Hristov, A.N., A. McAllister, F.H. Van Herk, K.J. Cheng, C.J. Newbold and C.J. Cheeke, 1999. Effect of *Yucca schidigera* on ruminal fermentation and nutrient digestion in heifers. J. Anim. Sci., 77: 2554-2563.

Hu, W., J. Liu, Y. Wu, Y. Guo and J. Ye, 2006. Effect of tea saponins on *in vitro* ruminal fermentation and growth performance of growing Boer goat. Arch. Anim. Nutr., 60: 89-97.

Jouany, J.P., 1996. Effect of rumen protozoa on nitrogen utilization by ruminants. J. Nutr., 126: 1335-1346.

Karsli, M.A. and J.R. Russel, 2001. Effects of some dietary factors on ruminal microbial protein synthesis. Turk. J. Vet. Anim. Sci., 25: 681-686.

NRC (National Research Council), 1984. Nutrient Requirements of Beef Cattle (6th Ed.). National Academy Press, Washington, DC.

Makkar, H.P.S. and K. Becker, 1997. Degradation of quillaja saponins by mixed culture of rumen microbes. Let. Appl. Microb., 25: 243-245.

Makkar, H.P.S. and X.B. Chen, 2004. Estimation of Microbial Protein Supply in Ruminants Using Urinary Purine Derivatives. Kluwer Academic Publishers.

Mao, H., J. Wang, Y. Zhou and J. Liu, 2010. Effects of addition of tea saponins and soybean oil on methane production, fermentation and microbial population in the rumen of growing lambs. Anim. Feed Sci. Tech., 129: 56-62.

Navas-Camacho, A., M.A. Laredo, A. Cuesta, H. Anzola and J.C. Leon, 1993. Effect of supplementation with a tree legum forage on rumen function. Livest. Res. Rural Develop. 5 (http:www.cipav.org.co/lrrd/lrrd5/navas.htm).

Odenyo, A.A., P.O. Osuji and O. Karanfil, 1997. Effect of multipurpose tree (MPT) supplements on ruminal ciliate protozoa. Anim. Feed Sci. Tech., 67: 169-180.

Patra, A.K. and Z. Yu, 2014. Effects of vanillin, quillaja saponin and essential oils on *in vitro* fermentation and protein-degrading microorganisms of the rumen. Appl. Microbiol. Biotechnol., 98: 897-905.

Santoso, B., A. Kilmaskosu and P. Sambodo, 2007. Effects of saponin from *Biophytum petersianum* Klotzsch on ruminal fermentation, microbial protein synthesis and nitrogen utilization in goats. Anim. Feed Sci. Tech., 137: 56-68.

Stern, M.D. and W.H. Hoover, 1979. Methods for determining and factors affecting rumen microbial protein synthesis: A review. J. Anim. Sci., 49: 1590-1602.

Suharti, S., D.A. Astuti, E. Wina and T. Toharmat, 2011. Rumen Microbial Population in the *In vitro* Fermentation of Different Ratios of Forage and Concentrate in the Presence of Whole Lerak (*Sapindus rarak*) Fruit Extract. Asian-Austr. J. Anim. Sci., 24: 1086-1178.

Thalib, A., Y. Widiawati, H. Hamid, D. Suherman and M. Sabrani, 1996. The effects of saponin from *Sapindus rarak* fruit on rumen microbes and performance of sheep. J. Ilmu Ternak dan Vet., 2: 17-20.

Wallace, R.J., N.R. Mcewan, F.M. Mcintosh, B. Teferedegne and C.J. Newbold, 2002. Natural products as manipulators of rumen fermentation. Asian-Austr. J. Anim. Sci., 15: 1458-1468.

Wang, Y.X., T.A. McAllister, L.J. Yanke, Z.J. Xu, P.R. Cheeke and K.J. Cheng, 2000. *In vitro* effects of steroidal saponins from Yucca Schidiigera extract on rumen microbial protein synthesis and ruminal fermentation. J. Sci. Food Agric., 80: 2114-2122.

Wina, E., 2005. The utilization of *Sapindus rarak* DC. Saponins to improve ruminant production through rumen manipulation. PhD dissertation, University of Hohenheim Verlag Grauer, Beuren, Stuttgart.

Wina, E., S. Muetzel and K. Becker, 2006. Effects of daily and interval feeding of *Sapindus rarak* saponins on protozoa, rumen fermentation parameters and digestibility in sheep. Asian-Aust. J. Anim. Sci., 19: 1580-1587.

Xu, M., M. Rinker, K.R. McLeod and D.L. Harmon, 2010. Yucca schidigera extract decreases *in vitro* methane production in a variety of forages and diets. Anim. Feed Sci. Tech., 159: 18-26.

Yost, W.M., J.E. Young, S.P. Schmidt and A.D. McGillard, 1977. Gluconeogenesis in ruminants: propionic acid production from a high-grain diet fed to cattle. J. Nutr., 107: 2036-2043.

Yuan, Z.P., C.M. Zhang, L. Zhou, C.X. Zou, Y.Q. Guo, W.T. Li, J.X. Liu and Y.M Wu, 2007. Inhibition of methanogenesis by tea saponin and tea saponin plus disodium fumarate in sheep. J. Anim. Feed Sci., 16: 560-565.