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Interaction Effects of Cattle Feed Supplement and Concentrate on Rumen Fermentability and Fiber Fraction Degradability in Low-Quality Forage

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Abstract: The objective of this study was to examine the interaction effect of additions cattle feed supplement (CFS) with feed concentrate on rumen fermentability and fiber degradability of low-quality tropical forages. The forage used is *Paspalum conjugatum* contain 5.44% of crude protein, feed concentrate contains 7.52% of crude protein and the CFS is containing 22.07% crude protein and 1.17% condensed tannin. The study was conducted *in vitro* using a split plot design with 2 main plots of full forage ration (FF) and forage + concentrate ration 60: 40% DM basis (FC), each plot was split with 4 levels of CFS (0, 5, 10 and 15% DM basis). The results showed the interaction effect of CFS addition and concentrate to crude fiber and fiber fraction degradation (P<0.05), but no interaction on rumen fermentability (P>0.05). The addition of CFS had a linear effect to increasing the rumen pH (P = 0.01), decreasing the VFA total (P = 0.03), decreasing the iso-VFA (P = 0.03) and decreasing the ratio of acetate / propionate (P=0.03). Dry matter degradability increased in CFS 10% (P <0.01), but not significant on organic matter degradability (P = 0.18). The interaction effect of CFS and concentrate was shown by decreased degradability of ADF (P = 0.03) and cellulose (P <0.01) in FF ration but an increase in FC ration and a higher linear increase to degradability of crude fiber, NDF and hemicellulose in FC than FF ration (P <0.01). The best, the CFS are given with feed concentrate at the level of 10%.
Key words : cattle feed supplement, degradability, fermentability, gambier, tannin.

INTRODUCTION

The productivity of beef cattle is very dependent on the quality of rations that are able to provide nutrients to maintenance and production needs. The ration of beef cattle are divided into forages, concentrates, and supplements. Forage is the main feed that is able to provide nutrients for cattle at low cost, concentrates can provide by high energy and protein sources, while supplements complement the nutrient deficiency.

Grass quality in the tropics including low-quality forages is characterized by low crude protein less than 7 g / kg DM and unbalanced nutritional elements [1]. In general, grass grows in coconut, rubber or oil palm plantations without maintenance, resulting in low quantity and quality [2]. Therefore it is necessary to feed concentrates from local feed ingredients such as sago, bran, cassava and coconut pulp to supply the energy and protein needed by cattle.

The low protein content in grass limits ruminal microorganisms to degradation of fiber forage [3, 4]. The use of low-quality forage for beef cattle feed requires nitrogen compounds to improve digestibility [5, 4] and starch feed ingredient to optimized rumen fermentability [6]. Ramaiyulis *et al.* [7] Have developed a Cattle Feed Supplement (CFS) containing soluble carbohydrates, nitrogen compounds, minerals and condensed tannins that can increase rumen microbial biomassa as rumen fermenters.

This research hypothesized that there is an interaction effect of CFS addition and feed concentrate on rumen fermentation and fiber degradation of low-quality forage. Our objective was to evaluate effects of increasing levels of CFS to fiber degradation low-quality forage with or without concentrate in the ration of beef cattle.

MATERIALS AND METHODS

Diet

Low-quality forage of tropical grasses of *Paspalum conjugatum* were used in this study. Concentrates are prepared from local resources mixtures of (% DM) 30% sago, 30% bran, 30% cassava, and 20% coconut pulp containing 11.39% crude protein and 75.20% total digestible nutrient.

Cattle feed supplement (CFS) are prepared by formula Ramaiyulis et al. (2016) mixtures of (% DM) 15% brown sugar, 27% bran, 15% soybean meal, 12% coconut meal, 15% tapioca, 5% urea, 3% salt, 3% mineral mix, 5% gambier leaf residue (*Uncaria gambir* Roxb). The study used a split plot design with 2 main plot of full forage ration (FF) and forage plus concentrate ration with ratio 60:40% DM (FC). Each main plot split with subplots are 4 levels of CFS (0, 5, 10, 15% DM ration). The nutritional content of the rations are shown in Table 1.

Table-1: Analyzed composition of diets

Item, %DM	FF[1]	FC[2]	CFS[3]
OM	90.33	90.86	85.95
CP	5.44	5.89	22.07
NDF	56.47	45.29	2.09
ADF	36.43	29.62	0.60
Cellulose	22.98	19.31	0.65
Lignin	10.86	7.50	-
CT[4]	-	-	1.17

[1]FF = Full forage without concentrate; [2]FC = Forage + concentrate (60:40% DM); [3]CFS = cattle feed supplement; [4]CT = Condensed tannin.

Experimental design and procedure

The study was conducted *in vitro* followed the first stage procedure of Tilley & Terry [8] using rumen fluid taken at animal slaughterhouse from cattle with low-quality tropical forage-based feed. Buffer solution [9] was mixed with rumen fluid at a ratio of 4:1 then filled to erlenmeyer flask 250 ml has contained 2.5 gr samples of treatment ration. Then flowed CO_2 gas for 30 seconds and incubated in shaker water bath at 39°C for 48 hours. Blanko is provided without sample and each treatment unit is repeated 4 times. Fermentation stopped by immersing the erlenmeyer flask in cool water for 30 minutes.

Then the contents of erlenmeyer flask measured with pH meters to determine ruminal pH and then centrifuged at a speed of 3,000 rpm for 5 minutes. The supernatant was used for the analysis of partial VFA concentrations using Gas Chromatography. The residue is washed with distilled water with centrifuged at the same speed and time for 3 times and then was filtered with Wathman 42 filter paper. The residue dried in an oven for analysis of dry matter and organic matter [10], neutral detergent fiber (NDF), acid detergent fiber (ADF), acid detergent lignin (ADL), hemicellulose (NDF-ADF) and cellulose (ADF-ADL) [11].

STATISTICAL ANALYSIS

The data were analyzed statistically using analysis of variance (ANOVA) and continued tests of simple effects, main effects and interaction effects and regression analysis to see the closeness of the relationship between factors [12].

RESULTS AND DISCUSSION

Table 2 shows no interaction effects of CFS and concentrate addition to pH, total and individual VFA (P> 0.05) in rumen fermentation of low-quality tropical forage incubated 48 hours *in vitro*. The ruminal pH was influenced by the addition of CFS which led to an increase in linear pH with an increase in CFS levels (P <0.01) in both FF and FC plots. The addition of concentrate lowered the pH value from 6.90 to 6.71 and with the CFS addition, the pH increased to 6.97 and 6.89 respectively for FF and FC plot at the 15% CFS level.

The effect of this increase in pH indicates that the CFS can reduce the pH drop due to the concentrate in the diet, this is due to the decrease in total VFA production due to the addition of CFS due to the decrease in fiber degradability by the condensed tannins contained in the CFS. Condensed tannins have no direct effect on rumen pH either *in vitro* or *in vivo* [13-15]. The pH range obtained in this study was 6.71-6.97 which is still at optimum pH for cellulolytic bacterial activity [16]. The activity of the cellulolytic bacteria will be inhibited if the pH of the rumen is below 6.0 [17].

The total VFA decreased linearly with the increase of CFS levels in both FF and FC plots (P <0.03), but no interaction effects of CFS and concentrate (P = 2.27) were found. The decrease in total VFA occurs at a CFS level of 10% with a decrease of 17.0% and 14.4% respectively on FF and FC plots. The decrease in total VFA production is due to the condensed tannin content in a CFS derived from the gambier leaf residue. The condensed tannin is an antinutrient that can form complexes with proteins, polymers (cellulose, hemicellulose, and pectin) and minerals that cannot be degraded by rumen microbial enzymes [19]. The decrease in total VFA was also reported as a result of the addition of chestnut tannin extract of 16.9% [18] and 11.7% on the addition of quebracho tannin extract [20].

Table-2: Interaction effects of CFS and concentrate on rumen fermentability and fiber fraction degradability

Item	Full Forage (FF)^{e}				Forage + Concentrate (FC)^{f}				SEM	P-value	
	CFS, %DM				CFS, %DM					L CFS^{g}	L CFS vs Con^{h}
	0	5	10	15	0	5	10	15			
Fermentability											
pH	6.90^{a}	6.90^{a}	6.94^{b}	6.97^{b}	6.71^{p}	6.78^{p}	6.84^{q}	6.89^{r}	0.01	0.01	0.61
Total VFA, mM	30.5^{b}	29.4^{b}	25.3^{a}	24.7^{a}	33.9^{q}	33.2^{q}	29.2^{p}	29.0^{p}	0.90	0.03	2.27
Acetate	21.9^{b}	20.1^{b}	16.4^{a}	16.2^{a}	22.2^{q}	23.4^{q}	17.2^{p}	17.3^{p}	1.59	0.04	5.52
Propionate	5.5^{a}	5.9^{a}	6.6^{b}	6.4^{b}	7.0^{p}	6.8^{p}	9.2^{q}	9.1^{q}	0.54	0.04	0.30
Butyrate	1.15	0.98	0.88	0.75	1.42	1.17	1.19	0.92	0.04	0.18	50.2
Valerate	0.90^{b}	0.27^{a}	0.28^{a}	0.25^{a}	1.07^{q}	0.35^{p}	0.34^{p}	0.32^{p}	0.18	0.03	4.61
Iso-VFA, mM	1.78^{b}	1.18^{a}	1.17^{a}	1.14^{a}	2.21^{q}	1.56^{p}	1.23^{p}	1.37^{p}	0.17	0.03	0.81
Iso-butirate	0.83^{b}	0.54^{a}	0.66^{a}	0.50^{a}	1.05^{q}	0.81^{p}	0.58^{p}	0.57^{p}	0.06	0.04	0.50
Iso-valerate	0.94^{b}	0.64^{a}	0.51^{a}	0.64^{a}	1.17^{q}	0.78^{p}	0.66^{p}	0.80^{p}	0.12	0.04	6.90
A/P	4.10^{b}	5.31^{b}	2.64^{a}	2.46^{a}	2.99^{q}	3.80^{q}	1.93^{p}	2.04^{p}	0.79	0.03	0.93
Degradability,%											
Dry Matter	49.0^{a}	49.7^{a}	51.9^{b}	51.6^{b}	57.2^{p}	58.7^{p}	61.1^{q}	61.4^{q}	0.48	<.01	0.14
Organic Matter	43.8	44.3	46.5	46.2	56.0	54.9	55.7	55.33	0.42	0.18	0.10
Crude Fiber	35.7^{a}	42.5^{b}	51.3^{d}	46.7^{c}	31.2^{p}	37.6^{q}	36.0^{q}	39.8^{r}	0.66	<.01	<.01
NDF	38.5^{a}	39.4^{a}	40.6^{b}	40.9^{b}	35.3^{p}	41.2^{q}	45.8^{r}	48.8^{s}	0.41	<.01	<.01
ADF	39.5^{b}	36.2^{a}	37.7^{a}	37.6^{a}	29.8^{p}	30.4^{p}	35.0^{q}	32.2^{q}	0.97	0.11	0.03
Hemicellulose	33.4^{a}	41.4^{b}	42.1^{b}	43.0^{b}	41.3^{p}	54.6^{q}	58.4^{qr}	64.8^{r}	2.07	<.01	<.01
Cellulose	46.4^{b}	39.9^{a}	42.1^{a}	43.0^{a}	41.2^{p}	41.4^{p}	44.7^{q}	47.6^{q}	1.72	0.09	<.01

^{abcd} = significant in FF plot; ^{pqrs} = significant in FC plot; ^{e} = plots of full forage without concentrate; ^{f} = plot of forage +concentrate in ratio 60:40% DM; ^{g} = linier effect of CFS; ^{h} = interaction effect of linier CFS and concentrate.

Regression analysis showed that there was a close relationship between total VFA production with degradation of NDF, ADF, hemicellulose and cellulose (P <0.1) with R^2 = 0.81, indicating that the total VFA produced in rumen, 81% from result of fibre fraction degradation (NDF, ADF, hemicellulose, and cellulose) and another 19% from other organic compounds such as proteins, fats, and vitamins. However, the relationship between total VFA and fiber degradability is partially contradictory because it is measured as VFA concentration in rumen fluid instead of VFA production in the rumen [21].

Increased the CFS levels resulted in decreased acetate production (P <0.04) and increased propionate production (P <0.04) and decreased acetate / propionate ratio (P <0.03). The condensed tannin content in CFS provides a change effect of VFA composition with increased propionate and decreased acetate as reported by Wischer *et al.* [14] that there is a decrease in acetate production compared to control by chestnut tannin treatment. The decrease in acetate production in plot FF is caused by decreased degradation of ADF and cellulose and increased degradation of NDF and hemicellulose due to contained condensed tannin compounds in the CFS. While in plot FC, the changes of acetate/propionate ratio are caused by depressed of fiber degradation and starch fermentation of the concentrate which yields high propionate. Accordingly, Cieslak *et al.* [22] Reported the addition of tannin *Vaccinium vitisidaea* 2 g / kg BK in dairy cattle ration with a forage: concentrate ratio 60:40 led to decreased acetate/propionate ratio.

Addition of CFS decreases the production of iso-VFA (P <0.03), the ie decrease of iso-butyrate and iso-valerate. Iso-valerate production has a pattern similar to the isomer of valeric acid. Iso-VFA can be used as an indicator of protein degradation occurring in the rumen [23]. Iso-VFA is produced from the degradation of protein derived branched amino acids, both derived from feed proteins as well as from microbial proteins. The presence of condensed tannin from gambier leaf residue in the CFS will bind some proteins to avoid degradation by rumen microbes as has been reported in previous studies [24]. Other researchers also reported that the addition of condensed tannin reduced the production of iso-VFA [20]. This mechanism has been used to protect proteins from rumen degradation to be available in the post-rumen gastrointestinal tract without modification of its constituent amino acids [25].

Dry matter degradability was influenced by CFS level (P <0.01), whereas organic matter degradability had no influenced by both treatment factor levels CFS or addition concentrat (P> 0.10). The dry matter degradability increases linearly and significantly at the CFS level 10% for both FF and FC plots.

There is an interaction effect of CFS level and addition of concentrate on the degradability of crude fiber in the rumen (P <0.01). In the plot FF, crude fiber degradability increased from control to 10% CFS level but decreased in 15%. In the plot FC, the addition of CFS with concentrate resulting linearly increasing crude fiber degradability with increasing level of CFS. Increased degradability of crude fiber was lower in plot FC with highest degradability of 39.8% obtained at 15% CFS level. Crude fiber degradability in plot FF decreased at the highest CFS level (15%) due to the condensed tannin resistance effect contained in the CFS to the crude fiber degradability. The higher levels of condensed tannins in the rumen will negatively affect the degradation of protein and fiber as a result of inhibition of growth or activity of cellulolytic microbes [26].

The interaction effects of additions CFS and concentrate was found on the degradability of the fiber fractions (NDF, ADF, hemicellulose, and cellulose) (P <0.01) as shown in Figure 1. Increased NDF degradability of forage in plot FF began to occur at the 10% CFS and non-significant with level l5%, whereas in plot FC there is a linear increase in NDF degradability with increased CFS level from 5% to15% (P<0.01). The CFS effect was higher than the feed concentrate effect on NDF degradability of 5.37% vs 2.89% and the CFS effect was higher in plot FC than plot FF of 4.6% and 0.8% respectively. NDF is a component of plant cell walls, higher contained in plot FF than plot FC. The rumen degradability of NDF is reduced by the addition of concentrate, due to decreased microbial fibrolitic enzyme activity attached to plant particles [28], as well as changes in the structure of the cellulolytic bacterial community [27].

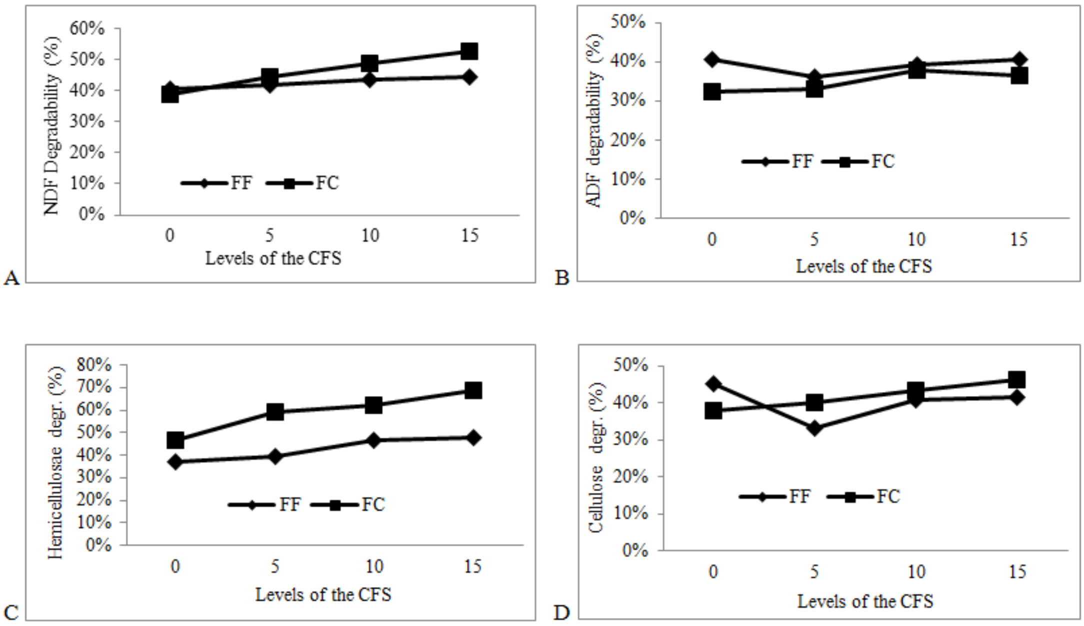

Fig-1: Interaction effects of the CFS dan the concentrate to fiber fraction degradability : (A) NDF, (B) ADF, (C) hemicellulose, (D) cellulose. The CFS levels (% DM ration). FF = full forage without concentrate; FC = forage + concentrate in ratio 40:60 %DM

The addition of CFS causes the decrease in ADF degradability in the FF plot and in otherwise an increase after the addition of concentrate (FC). ADF degradability is higher in FF than FC because ADF is a fiber fraction which contains high cellulose in FF plot. This study found that tannin has a negative effect on ADF and cellulose degradability but has a positive effect on NDF and hemicellulose degradability. Negative effects of tannin on the degradability of ADF and cellulose may be related to the formation of complex tannin ties with polymer cellulose polymers that cannot be degraded by rumen microbial enzymes [19] and a decrease in cellulolytic bacterial populations due to condensed tannin compounds [13].

The effect of CFS and concentrate on hemicellulose degradability shows a pattern similar to that of NDF degradability. The hemicellulose degradability in the FC plots increased linearly from 41.3% to 64.8% with CFS, whereas in the FF plot the increase occurred only at the 5% CFS level and did not differ significantly for the higher levels. The effect of concentrate addition on hemicellulose degradability was higher than CFS of 14.73% vs. 11.07% so that CFS addition gave higher benefit to increase hemicellulose degradability in FC plot.

Addition of CFS inhibited cellulose degradability in FF plot of 1.18% and showed a significant effect of decreasing cellulose degradability at CFS level of 5% (P<0.05). In FC plots, the concentrate increased cellulose degradability by 2.69% and showed significant at the CFS level of 10% (P<0.05). Tannins cause inhibition of fiber fraction degradation by microbes in the rumen because tannins form complex bonds with microbial enzymes [14]. Total populations of cellulolytic bacteria *Fibrobacter succinogenes* and *Ruminococcus* spp significantly decreased with tannin supplementation [19].

CONCLUSION

The addition of the CFS and the concentrate on low-quality tropical forage shows the interaction effect on rumen degradability of fiber fraction but not on pH, total VFA, and organic matter degradability. The tannin content in the CFS inhibits fiber degradability in the forage and with the addition of concentrates occurs the interactions that increase degradability linearly with CFS levels. addition the CFS should be performed with concentrate supplementation with the best level 10% on forage and concentrate ratio 60:40 (DM base).

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