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Exploring the Impact of Processed Cassava Peel on Microbial Dynamics and in vitro Nutrient Digestibility in Ruminant Diets

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ABSTRACT

This experiment aimed to explore the impact of increasing the amount of cassava peel in diets that had been soaked on whiting at different levels on rumen microbial biomass, protozoa populations, production of microbial protein, and *in vitro* nutrient digestibility. The research was arranged with a factorial pattern. There were two factors tested: the first was whiting dose and soaking time (0%, 3h; 0.25%, 3h and 0.50%, 2h), and the second was soaked cassava peel in diet (10, 20, and 30%). Diets were formulated to maintain a constant levels of protein and energy. The findings revealed that the increase of cassava peel in diets had no impact ($P>0.05$) on microbial biomass, protozoa populations, and microbial protein synthesis. However, soaking treatment with 0.25% whiting for 3h significantly ($P<0.05$) increased rumen microbial biomass. While nitrogen-free extract digestibility was only influenced by increasing cassava peel in ration. Fiber fraction digestibility was not different. In conclusion, using 30% cassava peel in the diet was safe for microbial activity and can support nutrients needed for growing rumen microbes.

Key words: Cassava peel, Invitro fermentation, Rumen microbial, Whiting.

INTRODUCTION

The rumen of animals has a large capacity as a place for the digestive process and feed fermentation. Rumen is inhabited by microbes consisting of anaerobic bacteria, protozoa, fungi, methanogenic archaea, and phages. They interact with each other and contribute, individually or in concert, to rumen function (Huws et al. 2018; Li et al. 2023). The dominant rumen microbial biomass are bacteria, protozoa, and fungi. The number of bacterial populations found in the rumen was 10^9-10^{10} cell/mL, while the protozoan population ranged from 10^5-10^6 cell/mL (Dijkstra et al. 2005). The protozoa population is estimated to be 50% of microbial biomass (Sylvester et al. 2009).

The ruminants nutrient requirement depends on rumen microorganisms since they are essential for digesting and producing volatile fatty acids (VFAs). VFAs are needed as the primary energy source for ruminants (Gebeyehu and Mekasha 2013; Ghimire et al. 2014; Pazla et al. 2021a).

Cassava peel is a feedstuff that has been proven to be energy source for dairy cattle (Agustin et al. 2020), but its use is still limited due to the limiting factor in its use, namely hydrogen cyanid (HCN). Efforts have been made to reduce HCN levels by soaking in calcium hydroxide (Agustin et al. 2021). By decreasing HCN levels, the amount of cassava peel in the diet is predicted to be increased. Cassava peel contains easily degradable carbohydrates, with a nitrogen-free extract content of 75.40%. According to Agustin et al. (2021), the highest VFA production (115mM) was found in soaking with a 0.50% whiting dose for 2h. Dijkstra et al. (2005) stated that the normal VFA concentration is between 70-130mM. During fermentation, energy is conserved in the form of adenosine triphosphate (ATP). ATP is used to maintain and grow the microbial population (Jamarun et al. 2017a). Thus, it is hoped that cassava peel in diets can produce the energy ruminants need. In addition, fermentable energy can increase N recycling to the rumen (Dijkstra et al. 2005). However, these easily degraded carbohydrates

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in high amounts will lower pH of the rumen that can affect the activity of cellulolytic microbes and also protozoa in the rumen.

Feed protein that escapes degradation in the rumen and microbial protein fulfils the protein requirements of ruminants (Church and Pond 1988; Pazla et al. 2023a; Zain et al. 2023). Ruminants can get their amino acids from microbial proteins. Microbial proteins contribute two-thirds of the amino acid requirements for ruminants. Factors influencing microbial protein production are the balance of nitrogen and carbohydrate sources (Pathak 2008; Jamarun et al. 2017b; Pazla et al. 2018; Pazla et al. 2021b). Trace mineral and vitamin deficiencies also affect microbial protein production. Hoover and Stokers (1991) stated that the rate of carbohydrate digestion influences microbial protein synthesis could be due to that the energy produced from carbohydrate fermentation is used to grow rumen microbes.

This experiment was designed and conducted to increase the use of cassava peel in a ruminant diet after reducing HCN levels through the soaking process and assess whether increasing cassava peel by 30% in the diet does not affect the activity of microbes in fermenting feed to produce energy and nutrients needed by ruminant animals. This can be seen from rumen microbial parameters in the form of microbial biomass, protozoa population, protein production from microbes, and digestion of nutrients. It is hoped that the results can provide the best results on rumen microbial biomass, protozoa population, microbial protein synthesis, and in vitro digestibility of nutrients.

MATERIALS AND METHODS

Ethical Approval

Ethical approval was not required as in this study live animals were not used.

Study Period and Experimental Site

From July to December 2022, this study was conducted at the Andalas University's Ruminant Laboratory of Animal Science Faculty.

Sample Preparation

The sample was cassava peel from a cassava chips manufacturing factory in Padang, Indonesia. Cassava peel contains nutrients as listed in Table 1. Cassava peel was cleaned, cut, and then soaked in lime water according to the length and treatment dose, namely 0, 0.25, and 0.50%. Cassava peel was then dried, ground, and used as a feed ingredient in diet formulations.

Research Implementation

The research was designed with a factorial pattern. There were two factors tested, i.e., the first was whiting dose and soaking time (0% 3h; 0.25% 3h, and 0.50% 2h), and the second was soaked cassava peel in diet (10, 20, and 30%). Diets were formulated to maintain constant level of protein and energy (Table 2). The ration was formulated using 50% forage and 50% concentrate.

In Vitro Method

The Tilley and Terry (1963) method was used to determine the rumen fermentation process and digestibility of nutrients in vitro. Each ration sample was weighed (2.5g) with 50mL of rumen liquid and 200mL of McDougall's solution in an Erlenmeyer flask. Anaerobic conditions were created by flowing CO_2 gas and incubated for 48h using a shaker water bath at 39°C. After 48 h, fermentation activity was stopped, and centrifuged. The residue was at the bottom, and the supernatant was on top. Residues were dried in an oven (60°C) after filtering through Whatman No. 41. Residues were used to determine nutrient contents using proximate analyses (AOAC 2016), nutrients' digestibility, and fiber fraction's digestibility (Van Soest 1994).

Table 1: Nutritive value of cassava peel before soaking

Nutrients	Nutritive value (%)
Organic matter	96.56
Crude protein	5.88
Total Digestible Nutrients	68.86
Ether extract	1.29
NFE	75.40
Crude fiber	13.99

Note: NFE = nitrogen-free extract

Determination of Protozoa Populations

The total protozoa population was determined using the method already described (Oghimoto and Imai 1980). These were computed using Neubauer chambers under a microscope. The rumen liquid was rotated at 3000rpm and placed at 4°C for 5min. A sample (1mL) of rumen fluid that has been incubated was mixed with methyl-green formalin saline (MFS). The filtrate results were dropped into the counting chamber as much as two drops and covered with a cover glass until evenly distributed. Used the counting chamber to know the protozoan population. The protozoan population was calculated using the formula:

Protozoan population (cells/ml) = 1x1000xCxFp / 0.1x0.0625x16x5

Note :

C: Number of colonies counted

Fp: Dilution factor

Quantification of Microbial Protein

Quantification of microbial protein was carried out using Lowry method (1951). Briefly, added 0.5mL of 1N NaOH, then boiled at 90°C for 10min. Prepared two tubes, and put 0.5mL sample in the first tube and 0.5mL distilled water in the second tube as a blank. Added 2.5mL of Lowry B solution to each tube, then homogenized and incubated for 10min, then 0.25mL of Lowry A solution was added to each tube, then again homogenized and incubated for 30min. The solution in both test tubes was read at a wavelength (λ) of 750nm. Protein contents were calculated by the equation:

$$Y = 0.0025X + 0.0146$$

Note: Y = production absorbance,x= protein content (µ/mL)

Determination of Microbial Biomass

It was calculated using the Griswold method (2003) to determine the amount of microbial biomass. Pipetted 1.5mL supernatant, centrifuged (15,000rpm) for 30min at

Table 2: Nutritive value of ration

Processed Cassava peel in ration	Nutritional value of treatment (%)								
	OM	CP	TDN	NFE	NDF	ADF	Cellu lose	Hemi Cellulose	HCN (ppm)
10% cassava peel, 0% whiting	89.69	10.58	64.57	53.59	59.10	30.95	23.13	28.15	2.69
20% cassava peel, 0% whiting	90.93	10.30	65.47	56.39	57.04	29.41	21.82	27.63	3.38
30% cassava peel, 0% whiting	92.17	10.00	66.38	59.13	54.99	27.87	20.52	27.12	8.07
10% cassava peel, 0.25% whiting	89.50	10.64	64.47	53.40	59.12	31.01	23.22	28.11	2.06
20% cassava peel, 0.25% whiting	90.55	10.39	65.28	55.99	57.10	29.53	22.01	27.57	4.13
30% cassava peel, 0.25% whiting	91.60	10.15	66.10	58.59	55.08	28.05	20.79	27.03	6.19
10% cassava peel, 0.50% whiting	89.58	10.62	63.14	53.28	59.35	31.38	23.43	27.97	2.60
20% cassava peel, 0.50% whiting	90.71	10.37	64.11	55.76	57.55	30.27	22.43	27.28	5.20
30% cassava peel, 0.50% whiting	91.83	10.11	65.07	58.23	55.75	29.16	21.43	26.59	7.80

Note: OM= organic matter; CP= crude protein; TDN= total digestible nutrients; NFE= nitrogen free extract; NDF= neutral detergent fiber; ADF= acid detergen fiber; HCN= hydrogen cyanid

4˚C. After that, it was washed using 85% NaCl solution and centrifuged again, dried in an oven at 60˚C for 48h and then weighed. The amount of microbial biomass was calculated by weighing the precipitate obtained.

Statistical Analysis

The data collected were analyzed using Analysis of Variance (ANOVA). Duncan's Multiple Range Test was used to know differences in effects between treatments (Gomez and Gomez 1984).

RESULTS

Rumen Microbial Biomass

The rumen microbial biomass (Table 3) of diets containing cassava peel processed at 0, 0.25, and 0.50% whiting 3, 3 and 2h was significantly (P<0.01) different, respectively. Using cassava treated by soaking with whiting significantly affected microbial biomass (P<0.01). However, the increase of using cassava peel did not affect microbial biomass (P>0.05). The highest (263.85mg/100mL) microbial biomass was found when cassava peel was soaked with 0.25% whiting for 3h while the lowest (189.63mg/100mL) amount of microbial biomass was found with 0.0% whiting for 3h.

Protozoa Population and Microbial Protein Synthesis

The protozoa population and microbial protein synthesis (Table 3) was not influenced (P>0.05) by treatments. No interaction between the two factors was detected. Treatment of cassava peel to reduce the HCN content also did not significantly (P>0.05) affect microbial protein synthesis (Table 3 and Fig.1).

Nutrients Digestibility

Nitrogen Free Extract (NFE) and Crude Fat Digestibility

The digestibility of nitrogen-free extract and crude fat from feed containing cassava was influenced by treatment. Increasing cassava peel use was highly significant (P<0.01) and affected NFE digestibility (Table 4). However, cassava treated by soaking with whiting did not significantly (P>0.05) affect NFE or crude fat digestibility.

Digestibility of Fiber Fraction

The fiber fraction digestibility consisting of neutral detergent fiber (NDF), acid detergent fiber (ADF), cellulose, and hemicellulose digestibility are shown in Table 5. The digestibility results for each fiber fraction were insignificant (P>0.05). When using 10% cassava peel in the ration, the digestibility value of neutral detergent fiber, acid detergent fiber, cellulose, and hemicellulose obtained was 61.67, 47.99, 60.67 and 77.54%, respectively. Increasing the use of cassava peel in rations to 30% obtained digestibility values for NDF, ADF, cellulose, and hemicellulose of 64.03, 54.66, 64.83, and 75.08%, respectively.

The effect of soaking cassava peel with whiting to reduce HCN levels was also not significantly different; it can be seen in Table 5. Fiber fraction digestibility was higher at soaked cassava peel in 0.25% calcium hydroxide, 3h. However, with the decrease in HCN levels, this digestibility value statistically did not differ (P>0.05).

Soaking treatment using 0.25% whiting with a soaking time of 3 hours generally provides a higher fiber fraction digestibility value. However, with the decrease in HCN levels, this digestibility value statistically does not show a significant (P>0.05) difference. Fiber fractions were higher at soaked cassava peel in 0.25% Ca $(OH)_2$, 3h.

DISCUSSION

Microbial Biomass

Rumen microbial biomass consists of bacteria, protozoa, fungi, amoebas and bacteriophage (Morgavi et al. 2010). Rumen microbes play an essential role in feed degradation in the rumen to produce energy for ruminants. About 70-85% of the energy needs for ruminants can be met from microbial biomass. Rumen microbes can also contribute 70-100% protein to ruminants (Thirumalesh and Krishnamoorthy 2013). In the present study, increase in microbial biomass could due to the soaking treatment of cassava peel with 0.25% $Ca(OH)_2$, 3h. Soaking cassava peel in $Ca(OH)_2$ damages the cell walls of cassava peel (Suismono and Prawirautama 1998), so enzymes produced by rumen microbes more easily digest it, thus in return provide energy for microbes to increase their growth and the amount of microbial biomass.

In the present study, there was also a decrease in HCN levels and the HCN value in diet was the lowest compared to other treatments. Rumen microbes have used the nutrients in rations containing cassava peel to meet the needs of microbial growth. Dietary HCN levels were decreased, but rumen microbial growth was unaffected. The increase in rumen microbial growth, characterized by

Table 3: The average rumen microbial biomass, protozoa population and microbial protein synthesis of cassava peel in diet and soaked in whiting

Processed cassava peel	Cassava peel in diet			Average
	10%	20%	30%	
Rumen Microbial Biomass (mg/100mL)				
0% whiting (3h)	175.55	186.67	206.67	189.63[c]
0.25% whiting (3h)	229.63	253.41	308.52	263.85[a]
0.50% whiting (2h)	199.26	202.60	191.11	197.78[b]
Average	201.48	214.35	235.43	
Protozoa Population (cell/mL)				
0% whiting (3h)	$2.69x10^6$	$1.78x10^6$	$1.77x10^6$	$2.08x10^6$
0.25% whiting (3h)	$2.90x10^6$	$2.15x10^6$	$1.95x10^6$	$2.33x10^6$
0.50% whiting (2h)	$2.15x10^6$	$2.08x10^6$	3.05x106	$2.43x10^6$
Average	$2.58x10^6$	$2.00x10^6$	$2.26x10^6$	
Microbial Protein Synthesis (mg/mL)				
0% whiting (3h)	19.38±1.37	19.01±1.39	18.50±2.13	18.96
0.25% whiting (3h)	16.48±0.95	18.59±2.30	20.74±0.06	18.60
0.50% whiting (2h)	20.67±0.11	20.19±0.97	18.62±0.53	19.82
Average	18.84	19.26	19.29	

Values (Mean+SD) bearing different alphabets in the same column showed a significant (P<0.01) difference.

Table 4: The average nitrogen-free extract and crude fat digestibility (%) of cassava peel in diet, soaked in whiting

Processed cassava peel	Cassava peel in diet			Average
	10%	20%	30%	
Nitrogen Free Extract Digestibility (%)				
0% whiting (3h)	69.72±4.71	71.20±5.46	70.17±4.63	70.36
0.25% whiting (3h)	70.81±5.04	69.80±6.78	73.14±4.09	71.25
0.50% whiting (2h)	62.13±7.16	70.88±5.19	72.66±3.89	68.56
Average	67.55[c]	70.63[b]	71.99[a]	
Crude Fat Digestibility (%)				
0% whiting (3h)	35.26±9.50	52.67±11.86	35.94±7.09	44.52
0.25% whiting (3h)	55.16±12.60	41.32±9.51	43.43±2.35	46.64
0.50% whiting (2h)	39.84±9.09	33.69±9.97	37.87±5.88	37.14
Average	46.75	42.56	39.08	

Values (Mean+SD) bearing different alphabets in the same row showed a significant (P<0.01) difference.

Table 5: The average NDF, ADF, cellulose, and hemicellulose digestibility (%)) of cassava peel in diet, soaked in whiting

Treated cassava peel	Cassava peel in diet			Average
	10%	20%	30%	
NDF Digestibility				
0% whiting, 3h	60.43±4.33	62.99±6.76	62.16±4.16	61.83
0.25% whiting, 3h	64.81±7.36	64.03±6.98	64.54±6.27	64.54
0.50% whiting, 2h	59.89±1.89	62.51±4.13	65.14±5.14	62.51
Average	61.67	63.17	64.03	
ADF Digestibility (%)				
0% whiting, 3h	46.99±7.56	52.08±5.41	51.01±4.68	50.28
0.25% whiting, 3h	53.98±4.55	52.78±2.38	55.40± 9.40	54.05
0.50% whiting, 2h	43.00±1.23	50.92±7.28	57.57± 4.93	50.5
Average	47.99	51.93	54.66	
Cellulose Digestibility (%)				
0% whiting, 3h	59.22±7.62	63.82±6.24	61.13±3.51	61.39
0.25% whiting, 3h	65.08±4.99	63.35±4.63	65.6±.61	64.68
0.50% whiting, 2h	57.71±4.17	66.65±2.71	67.76±1.53	64.04
Average	60.67	64.61	64.83	
Hemicellulose Digestibility (%)				
0% whiting, 3h	75.87±4.43	76.27±11.8	73.63±6.81	75.25
0.25% whiting, 3h	75.37±7.53	74.9± 2.72	75.96±5.93	75.4
0.50% whiting, 2h	81.37±5.47	72.87±11.29	75.65±7.82	76.63
Average	77.54	74.68	75.08	

There was no significant (P>0.05) effect of all treatments on fiber faction digestibility.

an increase in microbial biomass, will increase the number of enzymes produced to digest feed substances. Microbial biomass synthesis is determined by how much organic matter can be digested (Thirumalesh and Krishnamoorthy 2013).

Microbial activity increased, and the increase in the organic matter digestibility value of the ration proved this. This was supported by the results revealed by Suryadi et al. (2022). Rumen microbes play a role in producing hydrolytic enzymes to digest feed. Increasing the number of enzymes produced will increase the ability of the microbes to digest cassava peel. The nutrient needs of ruminants can be met from the nutrient content found in feed ingredients, which have undergone the digestion process in the rumen Putri et al. (2021). Processed cassava peels were more readily digested by the rumen microbes

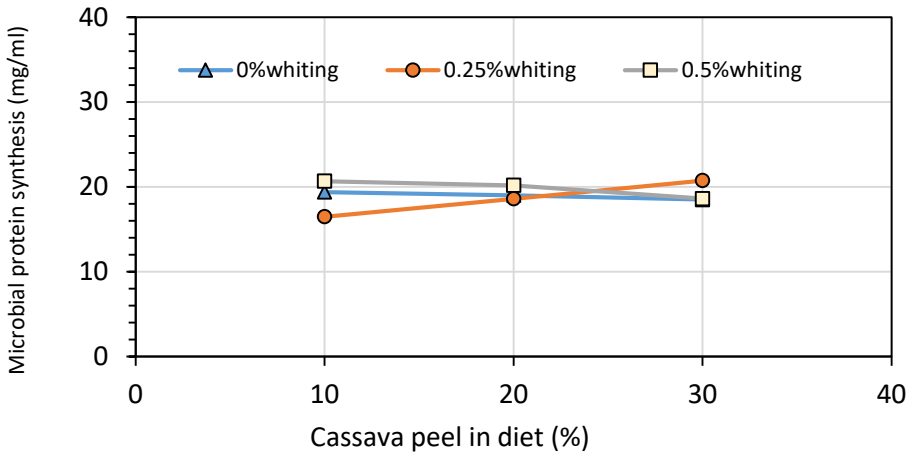

Fig. 1: Microbial protein synthesis of cassava peel in diet (10, 20, and 30%), soaked in whiting (0%, 3h; 0.25%, 3h; 0.50%, 2h).

and improved microbial biomass. The biomass found in our study was 175.55-308.52mg/100mL of rumen fluid. This value is higher than the results revealed by Ramaiyulis et al. (2019), namely 111.07-285.32mg/100mL in gambier leaf residue added in cattle feed supplement.

Protozoa Population

Protozoa contribute to the fermentation of carbohydrates in the rumen (Newbold et al. 2015; Aprilia et al. 2021), accounting for up to 50% of the biomass in the rumen (Sylvester et al. 2009). In our study, the protozoa population did not increase or decrease with the use of cassava peel in the ration up to 30% of the diet. HCN levels in the ration with increasing use of cassava peel to 30% also increased from 2.60 to 8.07ppm. Cyanide acid is toxic (Kutay et al. 2017; Gensa 2019), which can affect the metabolism of microbes in the rumen, including protozoa. When HCN is present in the rumen, it can disrupt this microbial balance by negatively affecting protozoa and other rumen microbes, particularly those responsible for fiber digestion. Based on the results obtained, it is proven that this dose of HCN is still safe for rumen microbial activity. Soaking the treatment of cassava peel with different levels and increasing its use in the ration did not interfere with the growth and population of protozoa. This means that the increase in microbial biomass found in this experiment was an increase in most bacteria because the protozoa population did not increase.

Based on the substrate, protozoa can be divided into protozoa that use soluble sugars, starch degraders, and lignocellulose degraders. The rations in this study were rich in starch with nitrogen-free extract content due to the increasing use of cassava peel in the diet. In starch-rich diets, protozoa can stabilize rumen pH (Jouany and Ushida 1999). So, indirectly, protozoa stimulate the activity of cellulolytic bacteria. Bacteria and protozoa work in degrading feed to provide the nutrients needed by the microbes and the ruminants themselves. The number of protozoa populations based on the use of cassava peel in rations ranges between $2.0x10^6$cell/mL up to $2.58x10^6$cell/mL, while based on the source of cassava peel used, the protozoa populations range between $2.08x10^6$cell/mL up to $2.43x10^6$cell/mL. The protozoan population obtained in this study was higher than the results revealed by Antonius et al. (2023). Dijkstra et al. (2005) stated that the rumen protozoa population of protozoa ranged from 10^5-10^6cell/mL, so the protozoa population obtained in our research was still in normal conditions for the rumen protozoa. Treatment of cassava peel to reduce its HCN content and its increasing use in rations does not disturb the protozoa population—an increase in the bacterial population results in an increase in feed fermentability. Protozoa maintain the concentration of fermentation products, including methane, ammonia, lactate, propionate, and butyrate (Pathak 2008).

Microbial Protein Synthesis

Factors that influence microbial protein synthesis are sources of nitrogen and carbohydrates, minerals, and vitamins available in the rumen (Pathak 2008; Pazla et al.2018; Putri et al. 2019). The source of protein for the ruminant animal comes from two sources, namely feed protein that has escaped rumen degradation and the second is protein derived from rumen microbes (Dijkstra et al. 2005; Pazla et al. 2023b). The differences in microbial protein synthesis are not significantly different due to the treated cassava peel. The main factors that influence this are the availability of nitrogen sources, carbon skeletons, and energy originating from the rations provided. In our study, energy availability in the form of VFA increased when using 30% cassava peel (Suryadi et al. 2022). The microbes in the rumen will use VFA as a carbon framework for protein production from microbes. These carbohydrate fermentation products, together with ammonia, will form microbial proteins (Karsli and Russel 2001). Increasing cassava peel in the ration resulted in decreased NH_3 levels (Suryadi et al. 2022). So, even though energy availability in VFA increases, it cannot increase microbial protein synthesis due to insufficient nitrogen sources.

Two dietary variables that have a significant impact on production of microbial protein are energy and protein availability. NH_3 in the rumen will be used to produce microbiological proteins. It is an indicator of the presence of the breakdown of protein entering the rumen. The growth and increase in rumen microbes depend on the availability of ammonia (Karsli and Russel 2001; Pazla et al.2021c; Suyitman et al. 2021). Treatment of cassava peel to reduce the HCN content also did not significantly affect microbial protein synthesis. We suggest that rations consisting of cassava peel need to be supplemented with a nitrogen or protein source. Compared with the results

revealed by Kardaya et al. (2010), the value of microbial protein synthesis in our study is higher, ranging from 16.48-20.74mg/mL. The efficiency of microbial protein synthesis is 14g Microbial Crude Protein/100g fermented organic matter. Microbial proteins contribute amino acids to ruminants. There are many factors that affect microbial protein synthesis, including synchronization of nitrogen and energy from the diet, rumen outflow rate/ rate of passage, minerals, and vitamins (Pathak 2008; Elihasridas et al.2023).

Nutrients Digestibility

Nitrogen Free Extract (NFE) Digestibility

Cassava peel contains easily digestible carbohydrates which can stimulate the growth and activity of protozoa in the rumen. This can lead to enhanced carbohydrate fermentation and increased volatile fatty acid production (Dijkstra et al. 2005). This is proven by the increased NFE digestibility value at a cassava peel percentage of 30% in the ration. This is due to the level of nitrogen-free extract in the ration also increasing from 53.40 to 59.19%, so the NFE digestibility value also increased, namely from 65.55 to 71.99%. It is known that nitrogen-free extract is an easily digestible carbohydrate. Cassava peel contains high levels of nitrogen-free extract, namely 75.40%.

Low HCN content will support the activity of rumen microorganisms in degrading feed components so that nitrogen-free extract digestibility increases, but this increase still produces results that are not significantly different. Karsli and Russel (2001) stated that feed fermentability increases with the microbial population.

Fiber Fraction Digestibility

The digestibility value of nutrients is primarily determined by the ability of rumen microbes to degrade the feed consumed by ruminant animals (Aprilia et al. 2021). The digestibility results for the fiber fraction (NDF, ADF, cellulose, hemicellulose) were not significantly different because microbial needs can be met by the presence of cassava in the ration, especially fiber-digesting microbes, including bacteria, protozoa, and fungi. The energy content in the form of TDN and crude protein in diets in this study were almost the same, so the availability of nutrients for microbes in each treatment was also almost the same. Treatment to reduce HCN levels by soaking cassava with different levels of whiting also showed results that were not significantly different. This is because the limiting factor for using cassava peel in rations in the form of HCN is still at a safe level, ranging from 2.06 to 8.07ppm; rumen microbes were still safe to carry out their activities.

Conclusion

Using 30% cassava peel in rations derived from soaking cassava peel with 0.25% whiting provides the best results on microbial biomass, microbial population, microbial protein synthesis, and nutrient digestibility. It can be stated that the use of 30% cassava peel in the ruminant diet, whose HCN levels have been reduced through a soaking process with 0.25%whiting for 3h, was save for microbial activity and can support nutrients needed for growing rumen microbes.

Authors Contributions

Fauzia Agustin designed the concept, searched for funding, conducted data analysis, and drafted and reviewed the paper. Roni Pazla and Novirman Jamarun supervised the laboratory work and draft the paper. Hannisa Suryadi prepared samples and conducted laboratory work.

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Conflict of Interest

The author declares that there is no conflict of interest.

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