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Condition Rumen Fluid Characteristics from Effects Indigenous Microorganisms in Fermentation Cocoa Waste

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Abstract:

Ruminansia animal feed ingredients consisting of forage consisting of grass and legume which are now her availability begins limited, to solve this problem do utilization agricultural wastes or wastes containing nutrient plantation low as alternative feed ingredients like cocoa rind. To overcome these problems required a simple technology that is fermentation using microorganisms indigenous so that the effluent quality chocolate can improve. Indigenous microorganisms are microbes that exist in individuals without disturbing activities. The purpose of this research is to improve the quality characteristics and digestibility of rumen fluid from the rind cocoa fermented with microorganisms indigenous. The method used in this study is completely randomized design factorial design with three replications, treatment factors consist of A is the level of microbes (A1 = 5%, A2 = 10%, A3 = 15%) and factor B is a long fermentation (B1 = 2 day, B2 = 4 days, B3 = 6 days) and the variables; the nutrient content and digestibility of cocoa rind before and after being fermented and the characteristics of rumen fluid in vitro methods. The results showed that increasing levels of use of microbes and fermentation time can improve skin quality cocoa beans that reduce the content of dry material from 15.691 to 13.634% and increase the protein content of the coarse 4.649 to 6.228%. Dry Material digestibility increased from 14.860 to 17.753% and Protein rough 40.472 to 54.519%. Rumen fluid characteristics are rumen fluid pH ranges from 6.427 to 6.603, NH₃ production berkisar 15,083 - 18,590mM, and Fatty Acid Volatile production of 97- 118.667 mg / 100ml. The conclusion that increasing the digestibility quality chocolate fruit leather can be made by fermentation by microorganisms indigenous.

Keywords: Fruit Leather Cocoa, fermentation, digestibility, microorganisms

1. Introduction

The main ingredient is a ruminant livestock forage food in general, which consists of grass and leguminosa which in the present availability is limited because the land began to grow grass and leguminosa already widely used for residential and business premises where food crops. Efforts to resolve this problem do the utilization of agricultural waste or agricultural waste products of low nutritional value as animal feed, the use of alternative feed ingredient does not change the ability of livestock production.

Waste food crops and plantations have an important role and potential in the supply of green feed for ruminansia livestock such as cattle, goats, sheep and buffalo, especially in the dry season. In addition, ruminant rearing system is still largely dependent on forage, feed in the form of grasses and other forage feed with little or no additional feed.

To overcome the problem of shortage of forage, farmers are expected to utilize agricultural wastes which are abundantly available in the vicinity include fruit skin cocoa, cocoa plant leaves, shoots of cane, rice straw, hay soybean and peanut hay through a certain treatment. Plantations in West Sumatra ± 82 450 ha. At the end of 2009, cocoa production is 40.250 tonnes (Prov West Sumatra Plantation Office, 2010), consisting of chocolate fruit rind of 73.73%, 2% of the placenta, and 24.20% seeds (Haryati and Hardjosuwito, 1984). Cocoa fruit skin is agro-industry waste produced cocoa plant (*Theobroma cacao* L). The results of proximate analysis cocoa rind contain dry matter (DM) 8%, coarse fibers (SK) 40.1%, Protein rough (PK) 8% and 50.8% TDN and Uses by ruminants 30-40% (IPPTP, 2001). To improve the nutritional value of the fruit skin cocoa with technologies such as physical treatment, chemical and biological, one of the technologies used are fermented. In the process of fermentation occurs solving complex compounds such as cellulose, hemicellulose, silica, by certain enzymes that reduce the content of crude fiber and improve the digestibility of the material. Fridarti research results (2013) that fermented fruit peels that use Basil sp chocolate can lower crude fiber

and increase the crude protein. Muzir study (2005) that the fruit skin cocoa fermented with yeast tape shows the physical shape and texture well, written by Izzati (2011) that fermented cocoa rind with MOL rind of chocolate can increase crude protein and lower crude fiber.

Indigenous microorganisms are microorganisms that exist in living things that do not interfere with her life. Several studies have isolation of microbial indigenous one by isolation and identification of bacteria of lactic acid and probiotic bacteria are generally investigated derived from fruits, animals and trash. Khairati (2011) have managed to characterize molecular Lactic Acid Bacteria from the cocoa-producing Bacteriocin green. Novianty research results (2011) also characterize molecular amylolytic lactic acid bacteria as probiotic potential of cocoa. Yunensi (2011) has managed to isolate and identify isolates of probiotic bacteria *Pediococcus pentosceus* origin cocoa. Utami, 2011. Successfully isolate and characterize Molecular Probiotic Bacteria (LAB) protiasse producer, lactase, fermenting green cocoa varieties.

The results show that the identification of the bacterial isolates of *Lactobacillus collinoides*. Isolation performed in MRS media (de Mann ROGOSA Sharp) were planted in the pour plate. The existence of these lactic acid bacteria in organic waste showed that the fermentation process that converts the cellulose in the waste into glucose by cellulase enzymes produced by the lactic acid bacteria.

2. Formulation of the problem

Is chocolate Fruit Leather digestibility can be improved by using microorganisms indigenous?

3. Research purposes

To Getting increase in the digestibility of nutrients from waste chocolate using microorganisms indigenous.

4. Research hypothesis

Microorganisms indigenous can increase digestibility Fruit Leather cocoa

5. Benefits of research

The results of this study can reduce the waste problem cocoa can be used as the building blocks for livestock rations ruminansia by breeders.

2. Materials and Methods

2.1. Material Research

The material used in the research is a microorganism indigenous derived from the rind of cocoa cultured on an agar medium and then made inoculum using fruit skin cocoa finely ground and then stored for 4 days, the results are used for the fermentation of fruit skin cocoa, research-scale labor using 100 grams of the fruit skin cocoa chopped and added to the level of microorganisms according to treatment (5, 10, 15%) and stored for 2, 4, 6 days, after it was dried in an oven with a temperature of 60°C so that the water content reaches 10 - 12% then analyzed proximate and in vitro.

2.2. Research Methodology

The design used was completely randomized design (CRD) factorial pattern of 3 x 3 with three replications, where factor A is the concentration of microorganisms (A1 = 5%, A2 = 10%, 15%), factor B is a long fermentation (B1 = 2 days, 4 days B2 =, B3 = 6 days). To determine the effect of treatment in early stages of the statistical analysis of variance. If there is a real effect of different treatments to do LSD (Steel and Torrie, 1995).

2.3. Implementation Research

1. Preparation Material cocoa rind cut into small pieces with a size of 2-5 cm.

2. Combine all ingredients in accordance with the treatment of each experiment / treatment input into plastic bags as a fermentation and tied up airtight.

Then treatment is stored according to treatment after which the samples were taken kelabor to be analyzed according to the variables.

2.4. Parameters Measured

The variables measured were:

1. Nutritional content and digestibility of the substrate before and after fermentation with the method of the proximate analysis and in vitro

2. Characteristics of rumen fluid and partial VFA Production

3. Results and Discussion

3.1. Mean Nutrient Content of Fermented Cocoa Rind

Results The mean study the nutritional content of fermented cocoa rind can be seen in Table 2. The dry matter content of fermented cocoa rind looks ranging from 13.634 to 15.691%. After statistically analyzed fingerprints manifold apparently, the result showed that the concentration factor and fermentation time and the interaction between the factors A and B show the different influences that were significantly ($P < 0.01\%$) to the average dry matter content of the fruit skin cocoa. The results of this study are lower than the results BPTP (2007) that fermented cocoa rind that use of probiotics containing approximately 18.4% dry matter. This result is also not much different from Fridarti study (2013) that uses *Basillus sp* in the fermentation of fruit skin cocoa is 15.28%.

No	Content	Factor A	Factor B (day)			mean
			B1 (2)	B2 (4)	B3 (6)	
1	Dry matter	A1	15,329 ^a	14,743 ^a	13,634 ^a	14,569 ^B
		A2	15,691 ^a	15,415 ^a	15,404 ^a	15,503 ^A
		A3	15,430 ^a	15,341 ^a	14,787 ^a	15,186 ^{AB}
		Mean	15,483 ^A	15,167 ^{AB}	14,608 ^B	
2	Crude Protein	A1	4,649 ^a	5,509 ^{a-c}	6,037 ^a	5,398
		A2	4,806 ^a	5,652 ^{a-c}	6,203 ^a	5,554
		A3	5,139 ^{a-d}	5,819 ^{a-b}	6,228 ^a	5,729
		Mean	4,865 ^a	5,660 ^a	6,156 ^a	
3	Crude fiber	A1	21,544	21,177	21,075	21,265
		A2	21,478	21,237	20,333	21,016
		A3	21,549	21,742	21,433	21,575
		Mean	21,523	21,385	20,947	
4	Crude lipid	A1	1,326	0,925	1,107	1,120
		A2	1,017	1,025	1,364	1,135
		A3	1,190	1,051	1,318	1,186
		Mean	1,178	1,000	1,263	

Description:
A, B, C = Average of the different superkrip a factor equal treatment for every different variables showed highly significant effect (P <0.01)
a, b, c = Mean superkrip different with the interaction between the same treatment for every different variables showed highly significant effect (P <0.01)

Table 2: Mean Dry matter content (DM), Crude Protein (CP), Crude Fiber (CF), Crude Lipid (CL), of Fruit Leather Cocoa Fermentation

If seen from the figures on the average dry matter research this is due present trend of increase in the water content due to the fermentation process resulting in a decrease of dry matter, it is suspected optimal capability of bacteria to digest the material feed, in accordance with the opinion of Solomon (1988) that the more the percentage of inoculum used the faster the fermentation process takes place due to the amount of fluid removed memalui metabolism will get more responses. Added by Fardiaz (1988) that mikroorganisme using karbohidrat after the first broken down into glucose. So, that the decrease in dry matter during the fermentation process due to a respiration of the bacteria that produce the energy, water and carbon dioxide. Microorganisms decompose organic matter and remains alive into the elements simpler.

Mean crude protein content fermented fruit skin cocoa is seen ranging from 4.649 to 6.228%, after a statistical analysis turned out to be the result of variance showed that the fermentation time factors and the interaction of both factors memeperlihatkan their highly significant effect (P > 0.01%) on the average Crude protein content, while the concentration factor adanyan not show no significant effect (P <0.05%) of the crude protein kandungn cocoa rind. The results of this study are not much different from the results of research BPTP (2010) fermenting with yeast cocoa rind. It is presumed by increasing the concentration can increase the number of bacterial cells and the amount of enzyme that is produced as a result of the activity of these bacteria as well as the addition of stool chicken where feces chicken has nutritional value quite well as animal feed and high in protein and serves to complement the food substances such as carbon, nitrogen necessary for growth and proliferation of microbes. This is in accordance with the opinion of Sukara and Admowidjojo (1990) that the growth and proliferation of microorganisms has good and can turn more components of the media into a mass of cells, resulting in the formation of proteins derived from microbial body itself will increase the crude protein. Added by Buckle (1987), that the fermentation is generally result in partial loss karbohidrat of food, but the loss was covered by the gains that protein.

On the average of crude fiber content of fermented cocoa rind looks ranging from 20.333 to 21.742%. Having analyzed statistically apparently, the result of variance to the average content of crude fiber cocoa rind showed no significant effect (P > 0.05) at factor A, factor B and the interaction between factors A and B. If viewed as figures on the average research Extra mikrooranisme concentration to be down at the level of 10% but rising back to the level of 15%. Research results obtained are not much different from the results BPTP West Sumatra (2010) that fermented cocoa rind that uses Prebiotic Fiber contains roughly 24.7%. This is presumably related to the growth and proliferation of bacteria. Opinions according Sulaiman (1988) that the dose of inoculum is a very important factor in the fermentation inoculum dose where certain environments and certain fermentation time anyway. Written by Herman (2011) that Fermentation is one of the technologies to improve the nutritional value of feed high in fiber. Fermentation can hydrolyze the protein, fat, cellulose, lignin and other polysaccharides, so that the fermented material will have a higher digestibility, but it also increased the crude protein of these materials.

3.2. Analysis of Van Soest

Van soest (1982) states that the content of ADF (acid Detergent Fiber) in the ration may decrease the digestibility of nutrients. From Table 3 shows that fermentation with microorganisms indigenous able to increase NDF fermented cocoa rind.

Komponen (%)	Cocoa rind	Cocoa rind Fermentation
Netral Deterjen Fiber (NDF)	56,45	59,68
Acid Deterjen Fiber (ADF)	48,86	50,77
Hemicellulose	6,12	8,91
Cellulose	21,76	21,70
Lignin	27,39	27,27
Silica	1,82	1,80

Table 3: Chemical Composition cocoa rind and cocoa rind fermentation

Impairment analysis column fermented rind cocoa prove that the action of the enzyme produced has started to break down the lignin and silica. As a result, crude fiber contained in the leaves cocoa and cocoa rind decreased. Hai is suspected as the cause of the cellulose and hemicellulose which binds to lignin has been converted into simple sugars.

2. Digestibility Dry Materials (BK), Protei Coarse (PK), Crude Fat (LK), Crude Fiber (SK) and Rumen Fluid Characteristics of Leather Cocoa Fermentation

The mean digestibility fermented cocoa rind can be seen in Table 5, the average digestibility Ingredients Dried rind cocoa ranging from 14.860 to 17.882%.

Tabel.5. The mean digestibility Dry Materials (DM), Crude Fat (CL), Crude Protein (CP), Crude Fiber (CF) and Rumen Fluid Characteristics of fermented cocoa rind

No	Digestibility	Factor A (The concentration of Mikrobes)	Factor B (Fermentation)			Mean
			B1 (2 days)	B2 (4 days)	B3 (6 days)	
1	Dry matter	A1 (5%)	16,891 ^{bc}	15,193 ^c	14,860 ^c	18,189 ^c
		A2(10%)	17,303 ^{abc}	16,298 ^d	16,571 ^{cd}	19,488 ^b
		A3(15%)	17,753 ^{ab}	17,882 ^a	17,251 ^{abcd}	20,580 ^a
		Mean	17,316 ^a	16,458 ^b	16,228 ^b	
2	Crude protein	A1 (5%)	40,472 ^c	47,790 ^c	54,519 ^a	47,594 ^b
		A2(10%)	41,655 ^{bc}	48,325 ^{bc}	54,029 ^a	48,003 ^{AB}
		A3(15%)	42,205 ^d	49,486 ^a	53,889 ^a	48,527 ^a
		Mean	41,444 ^c	48,534 ^b	54,146 ^a	
3	Crude protein	A1 (5%)	20,730 ^{bc}	21,407 ^{cd}	20,113 ^c	20,750 ^b
		A2(10%)	21,806 ^{bc}	22,252 ^{ab}	21,036 ^{cd}	21,698 ^a
		A3(15%)	21,938 ^{bc}	22,352 ^a	21,949 ^{bc}	22,079 ^a
		Mean	21,491 ^{AB}	22,004 ^a	21,033 ^b	
4	Crude lipid	A1 (5%)	20,265 ^a	9,961 ^c	11,838 ^c	14,022 ^b
		A2(10%)	21,348 ^a	16,193 ^b	16,518 ^b	18,020 ^a
		A3(15%)	21,032 ^a	14,796 ^c	19,889 ^a	18,572 ^a
		Mean	20,882 ^a	13,650 ^c	16,082 ^b	
5	Organic materials	A1 (5%)	81,467 ^d	79,157 ^d	86,470 ^c	82,365 ^b
		A2(10%)	85,267 ^c	86,771 ^c	91,584 ^b	87,874 ^a
		A3(15%)	85,175 ^c	86,634 ^c	95,727 ^a	89,179 ^a
		Mean	83,970 ^b	84,187 ^b	91,260 ^a	
6	BETN	A1 (5%)	40,995 ^{ab}	31,368 ^d	31,951 ^d	34,771 ^b
		A2(10%)	43,612 ^a	38,446 ^{bc}	37,554 ^c	39,871 ^a
		A3(15%)	42,970 ^a	37,148 ^c	41,838 ^a	40,652 ^a
		Mean	42,526 ^a	35,654 ^b	37,115 ^b	
charakteristics of rumen fluid						
5	N-NH ₃	A1 (5%)	15,627	17,990	16,547	16,721
		A2(10%)	18,767	16,997	16,963	17,576
		A3(15%)	15,083	18,590	16,783	16,819
		Mean	16,492	17,859	16,764	
6	VFA	A1 (5%)	118,667	99,000	101,000	106,222
		A2(10%)	99,667	100,667	109,333	103,222
		A3(15%)	108,667	103,333	97,667	103,222
		Mean	109,000	101,000	102,667	
7	pH	A1 (5%)	6,480	6,527	6,520	6,614
		A2(10%)	6,427	6,493	6,603	6,552
		A3(15%)	6,540	6,563	6,560	6,561
		Rataan	6,482	6,528	6,561	

Description: Value superscript on the line and show the influence of different colom highly significant (P < 0.01)

Table 4

Dry matter after statistically analyzed the results of variance stated that their significantly different between treatments ($P < 0.01$). The average obtained from this study at 15% concentration of microorganisms and fermentation time 4 days highest (17.882%) and the lowest concentration found in the treatment of 5% to 6 days' fermentation time (14.860%). This shows that the concentration of 5% of microorganisms with 6 days' fermentation time is a good time to breed sufficient sources of nutrients. It is also not too much different from the results of research Atmadja (2008) that fermented cocoa rind using *Trichoderma harzianum*, get the average dry matter fermented cocoa rind highest concentration of 4% to 8 days' fermentation time.

The mean digestibility Protein Cocoa Fruit Fermentation Rough skin can be seen in Table 4, ranging from 40.472 to 54.519 after statistically analyzed the results of variance stating that the concentration and length of fermentation as well as the interaction between the different influences that were significantly ($P < 0.01$) the digestibility of fermented cocoa rind. Results of research concentration is highest at 5% to 6 days' fermentation time (54.519%) and the lowest at 5% concentration treatment with fermentation time 2 days (40.472%).

The average content of VFA and NH_3 inversely related research results, the average content of VFA looks ranging from 15.083 to 18.767 mM while NH_3 visible from 97.667 - 118,667.mg / 100ml. Statistical test results analysis of variance directing that the concentration factor, long fermentation and the interaction between the two factors had no significant influence ($P > 0.05$) against the VFA content of fermented cocoa rind. The results of this research have been to meet the needs of the rumen microbes to grow and thrive but rumen microbial activity is still low. Randahnya rumen microbial activity can lead to substance digestibility meal will go low. VFA content of this research is already disqualify pertumbhan and rumen microbial activity. This is in accordance with the opinion of Sutardi (1979) that the VFA concentration in rumen fluid needed for growth and microbial activity is 80-160 mM.

Fermentasi on time	As Asetat	As Propionat	As Iso Butirat	As n Butirat	As Iso Valerat	As n Valerat
2 days	36,26	16,42	3,91	5,28	1,93	2,58
4 days	37,22	18,51	4,26	9,77	2,27	2,55
6 days	41,05	20,04	4,67	6,82	2,53	2,83

Table 5: Results of analysis of VFA Partial lather cocoa fruit with a concentration of 15% fermentation Microorganisms indigenus

Mean rumen fluid pH studies ranged from 6.427 to 6.603. Statistical Test Results of analysis of variance showed that the concentration and length of fermentation cocoa rind gives no significant effect ($P > 0.05$) to the average pH of rumen fluid. In figures rumen fluid pH increases with increasing concentrations of microorganisms and fermentation time. This is in accordance with the opinion of Owens and Bergen (1983) To be able to grow optimally and run metabolic processes, rumen microbes require optimum environment that is pH ranging from 5.7 to 7.2 and a temperature of 38-41°C.

Rumen fluid state influenced rumen fluid pH value is the length of time the food was backdated to stay fed and salivary secretion. Saliva is a bicarbonate buffer around 100mm, which is available to neutralize the continuous production and increase during the meal and ruminas. Maximum fermentation in ruminants occurred five hours after eating. At 4-5 hours after a meal, rumen pH is relatively neutral (ideal conditions) to ensure maximum process because the pH value is lower at the time of 0.5-4 hours after a meal, then be balanced, because the acid production and incoming buffer of saliva or bases of feed. Guaranteeing maximum process because the pH value is lower at the time of 0.5-4 hours after a meal, then be balanced, because the acid production and incoming buffer of saliva or base of the feed.

4. Conclusions and Recommendations

From the research results, can be concluded as follows:

1. Microorganisms indigenus concentration of 15% with a 6-day fermentation time can increase the protein content Rude, Crude Fat, NH_3 and lower content of dry material, Crude Fiber, VFA cocoa rind
2. The concentration of microorganisms indigenus of 15% can improve the digestibility of Dry Ingredients, Protein Rough, Rough Fat, Crude Fiber, NH_3 , VFA, and BETN cocoa rind, while Fermentation time can decrease the digestibility of cocoa rind

4.1. Suggestion

1. It should further study the addition of other ingredients such as minerals added to the fermentation process to increase the activity mikrooragisme indigenus cocoa rind of making it more effective and optimum.
2. It should further study the use of increased levels of cocoa rind fermentation in livestock rations ruminansia sheep and other livestock.

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