Comparison of isolate dadih with yeast dadih in improving nutrition quality of Cassava Waste (CW)

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Abstract. The cassava industry in North Sumatra Province was one of the most significant agricultural industries. Waste from the cassava industry which was called cassava waste/CW/Onggok was used as feed for ruminants such as cattle, sheep and monogastric such as pigs. The low nutrients in CW caused the need to find a way for improving the nutrients quality. This research was conducted with the aim to help livestockers to ferment their livestock feed. This study compared the ability of fermentation between dadih isolate with dadih yeast. Dadih is traditional food in Indonesia where milk is fermented in bamboo tube. Dadih yeast was made by mixing dadih and whey with flour, made in around shape and sun dried. The results showed that pH of CW by dadih isolate was the lowest while crude protein, crude fiber and fat in CW treated with dadih isolate were improved significantly compared either to control or to dadih starter while fermented CW was better than non-fermented CW. It was recommended livestockers to ferment CW by using either by dadih isolate or dadih starter.

Keywords: dadih isolate, dadih starter, CW, nutrition quality

1. Introduction
In 2015, the province of North Sumatera was rated as the second biggest cassava producer in Sumatera together with Lampung [1]. Cassava production was 1,619,405 ton which mostly processed into tapioca starch by many big tapioca factories in North Sumatera Province and the raw materials are supplied by perkebunan ubi masyarakat from kabupaten yang berdekatan.

There were several regencies that significantly produced cassava, namely Serdang Bedagai, Simalungun, Deli Serdang, Tapanuli Utara and Tapanuli Tengah. Cassava production were 447,990 tons, 380,701 tons, 178,790 tons, 64,853 tons, 33,648 tons respectively in 2014 [2]. There were other regencies that also produced cassava however as they produced only small number, usually tapioca produced locally in small factories.

From the cassava industry were produced abundant cassava waste (CW) because for every ton of cassava were produced 114 kg waste. In North Sumatera, CW was used for animal feed, such as beef cattle, sheep, pigs and poultry. Livestock industry in North Sumatera generally utilizes CW in dry conditions that were ordered directly from tapioca factories. For small-scale livestockers generally they buy CW in wet conditions. On the edge of the road in North Sumatera, it could be found pile of CW sell. Small livestockers who used CW for feed often complained of slow livestock production. This was due to the low nutrient content of the CW which was estimated to be only able to meet the basic needs of livestock life, so for growth, pregnancy and lactation required additional feed to meet protein and energy needs of livestock [3].

In North Sumatera, livestockers community have been familiar with the application of fermentation feeding. Fermentation was a very old way/thousand years ago which supported higher final
concentration of products with environmental advantages [4]. Until now the simple fermentation technique in North Sumatera that was socialized to livestockers was to utilize the yeast of traditional food which called tape which contained *Saccharomyces cerevisiae* on CW. But some breeders have the assumption that fermentation with yeast tape could cause reproductive disorders. CW could be hosted by microbial diversity and the true microbial would support effective utilization of CW [5]. Therefore, it is necessary to find more kind microorganisms especially those which could applied by simple fermentation and especially those microorganism that could easily to have.

This study aims to investigate alternative microorganism for CW fermentation. The result would help the livestock industry, i.e (1). producer which were CW factories (2). user which were livestockers. Microorganisms were isolated from dadih which is a traditional food in Indonesia. Livestock milk either buffalo or goat was fermented in bamboo tube. After 2 or 3 days dadih was emerged which contained of 36 strain of lactic bacteria [6] and was dominated by *Lactobacillus plantarum* [7] while [5] found *Bacillus licheniformis* and *Bacillus subtilis* were the dominant bacterial species in cassava peel waste, *Lactobacillus fermentum* and *Lactobacillus plantarum* were the dominant bacterial species in liquid CW. *L plantarum* was also found in liquid CW [8]. It was assumed that by using BAL from dadih, either CW producer or CW user will be greatly get benefit because bamboo could be found in the whole area of Indonesia up to a very remote area while milk also was everywhere either fresh milk especially commercial milk/dry milk. The output of this study was also that livestockers in rural area could do fermentation without depend on commercial fermentor.

2. Materials and Methods

This research started with making dadih which was a process of fermentation by entering goat milk into bamboo tube. The development of microorganisms spontaneously was occured that was dominated by lactic acid bacteria (BAL) although there were also mold and yeast. The fermentation was occured within 2 days. Furthermore, in this research, the isolation of microorganisms from fermented whey was conducted. Next, the preparation of yeast dadih which was made by mixing sticky rice flour, dadih and whey then left alone for 1 night. The next day the dough was formed into a sphere weighing 10 grams and dried in the sun to dry.

The identification of microorganisms only focused on lactic acid bacteria through the slide culture method on microorganisms growing on PDA medium which only selected 3 directly solitary. The next three selected BALs were developed and processed into bioactivators which be used to ferment CW. Fermentation was done by steaming CW. CW that was cold then put in tupperwares each of 5 replication. The research method was used Completely Randomized Design (RAL). Fermentation was carried out for 10 days. Treatments were as follows: T1 (without bioactivator), T2 (bioactivator BAL), T3 (curd yeast bioactivator). Parameters of research were pH, crude protein, crude fiber and fat.

3. Results and Discussions

In this study fermented CW was dominated by lactic acid bacteria. This was consistent with Sunarlim et.al [7] that on dadih was found BAL which was dominated by *Lactobacillus plantarum*. In the fermentation of materials that were inoculated by microbes would undergo changes in their chemical composition. Fermentation of carbohydrate-containing substances would produce alcohol. In this study also found the smell of alcohol. By Sunarlim [7] it was stated that any particular fermentation process was caused by certain microbial activity, which was specific to the fermentation process. An example lactic acid fermentation by *Lactobacillus* bacteria. The working bacteria had their own features that were related to the benefits it provides to the environment. In this study observations were on pH, crude protein, fiber and fat.
Table 1. pH and nutrient content of CW and fermented CW by isolat dadih and yeast dadih

|            | pH  | Crude Protein | Crude Fiber | Fat  |
|------------|-----|---------------|-------------|------|
| CW         | 4.8 | 2.28 C        | 20.23 B     | 0.68 C |
| CW + Isolate | 3.4 | 7.48 B        | 17.86 A     | 2.64 A |
| CW + Yeast | 4.1 | 4.94 A        | 17.59 A     | 1.26 B |

3.1. pH

From Table 1 it was known that there was a decrease in pH, initial pH 4.8 to 3.4 and 4.1 in line with increasing fermentation time. Araujo [9] fermented with BAL from Youghurt found a decrease in pH between 3.84 and 3.98. According to Sunarlim [7] dadih were dominated by BAL especially *Lactobacillus plantarum*. *Lactobacillus* was a class of lactic acid-producing bacteria, including gram-positive bacteria, anaerobic faculty that had two mechanisms to obtain energy. When there was oxygen, energy was obtained by aerobic respiration, when no energy oxygen was obtained by anaerobic fermentation. Lactobacillus in animal feed materials was an indication of a healthy environment because these bacteria were normal microflora in the environment and in the digestive tract of living things. The resulting lactic acid will inhibit various pathogenic microorganisms. In the feed industry *lactobacillus* bacteria was widely used as a probiotic directly applied as a mixture of animal feed. For the feed industry, the decreasing pH indicates that the feed material was in good condition because the acidic pH causes the decay bacteria to be inhibited. Some other benefits of *Lactobacillus plantarum* were inhibiting pathogenic bacteria, improving immunity, can produce vitamin B and bacteriocin, antimutagenic, anticarcinogenic [6].

For ruminants, feed that has an acid pH is not a problem because when the cattle chew the feed, saliva of cattle that has an alkaline pH causes the process of neutralizing the feed ingredients. Observations made by Ginting on fermentation CW application with BAL-containing bioactivators on Barbados Blackbelly sheep, fermented CW for 2 months remained palatable.

3.2. Protein

On livestock feed, conventional sources of protein were very expensive, for example fish meal and also inadequate. This was why, efforts to find nonconventional sources were of much concern, for example through the state of fermentation. In this research, protein was increased significantly from 2.28 (control) into 7.48 (isolate dadih) and 4.94 (yeast dadih). It was assumed that isolate dadih contained lots more microorganisms than in yeast dadih thus was followed by enzymatic-pools. This was in concordance with Soccol[4] which mentioned that fermentation was how to produce enzymatic-pools which mainly proteases, amylases and lipases to hydrolyzed sub-strate macromolecules like starch, proteins, fatty acids. Aruna et.al [10] found an increased on protein in yam after 96 h, using Saccharomyces cerevisiae (BY4743). Samples from fermented biomass were collected at 24 h interval for 96 h, oven dried at 60 C, cooled, milled and assayed for chemical and amino acid composition. The crude protein, true protein, fat and ash contents increased significantly from 6.60%, 4.38%, 1.12% and 4.45% to 15.54%, 13.37%, 2.09% and 8.02% respectively when fermentation reached 96 h in the presence of ammonium sulphate. Any differences in nutrient quality after fermentation were depended on most suitable microorganisms with substrated, nutrient content of substrates and treatment addition [11]. However, any kind of microorganisms had their own specific good, for example *L. plantarum* which could be acted as probiotic. Also, the most important thing was the technology could be easily be implemented in field, easily to be found and cheap.
3.3. Fiber
Crude fiber from CW control was about 20.23% and there was a decreased about 3% due to fermentation. The initial crude fiber depends on the milling process taking place at tapioca factory. In North Sumatera, generally the new generation machines cause crude fiber smaller in CW. Fiber in this study became better about 3%. This was probably because BAL did not produce cellulase enzymes to degrade fibers. Actually the fiber in this study was about 17% which good enough for ruminants [3]. That was why feedlot industry of beef cattle found in North Sumatera generally use CW as animal feed material. The need for feedlot CW in North Sumatra was very large. In addition to feedlot, pig farmers for the Simalungun and Toba areas also use CW. For example in North Sumatra there were 6 feedlot/cow feed industries, among others, Lembu Andalan Langkat Enterprises that maintained 3500 cows. Daily dry daily needs of about 10,500 kg because livestock feed everyday were 3,500 x 10 kg = 35,000 kg and CW was 30% or about 300 ton dry CW every month. In North Sumatera, based on BPS [2], there were 184,612 ton of CW and which were predicted not enough to supply feedlot industries. That was why, part of CW supply came from out of North Sumatra which was Lampung. Only small livestockers in North Sumatra were used mostly local CW.

3.4. Fat
The fat in this study differed significantly between controls by treatment of isolate dadih and with whey yeast dadih. This was occured because BAL microorganisms that work to remodel starch in CW result in increased fat from the activation of the lipase enzyme [4]. According to Ghaffar et al [12] BAL made eco-friendly processing and BAL also had a fermentable capability on many of the agricultural and agro-industrial by-products. BAL became attractive candidates in fermentation biotechnology to produce a value added product with multiple applications.

3.5. Utilization of BAL Dadih, strategic effort to improve the quality of animal feed ingredients
As mentioned before, the result of this research will help livestockers industry. In Figure 1 it can be seen the process in tapioca industry. CW was generated after the pressing process. The water content of the pressing depends on the machine type in the cassava factory. However for fermenting process the water content of CW should be about 60%. In the interest of raising the quality of CW then some strategies can be made by utilizing the results of this study. For feedlot industries that require dry CW, CW fermentation became difficult due to space problems. CW should be fermented at least 4 days [4] while the volume of CW were so enormous that the factory must invest to build a fermentation room. Therefore the most feasible CW was implemented for non dried CW. In Indonesia, the factory directly packs the CW into a plastic sack.

In cassava factory, when CW coming out of the exhaust pressing pipe, the application of microorganisms can be done by spraying bioactivator on CW. Automatically microorganisms will develop inside the plastic sack because BAL was anaerobic facultative bacteria. Usually about 2 months time to spend stock of CW sell. Ginting discovers from unpublished studies that up to 2 months CW fermentation still in good condition and favored by livestock. CW purchased by small livestockers can also be fermented alone by livestockers by utilizing yeast dadih. With yeast dadih that can be made by livestockers themselves, they can save time, and bamboo plants become sustainable because livestockers do not need to cut bamboo everytime they need dadih. Livestockers just need to open the CW plastic sack a little for dadih yeast entry, flatten yeast and close the plastic sack for a fermentation process.

The advantages of dadih microorganisms because these microorganisms can be developed very easily through milk and bamboo tube. People even who live in very remote area which usually cannot have
biofermentor can buy available commercial milk powder, pasteurized it, process it in bamboo tube and after 2 or 3 days become dadih.

Furthermore, the dadih was processed into dadih yeast by mixing the dadih with glutinous flour (250 g) mixed with cornstarch (50 g), dadih 30 g, 230 cc whey left to stand for 1 night, rounded and dried to dry. Preliminary research conducted by Ginting by using rice flour, cornstarch and glutinous flour, then the best result was to use glutinous starch mixed with cornstarch. If people already have whey yeast, people can keep it in the refrigerator to keep it fresh. Thus the community does not need to sacrifice many bamboo trees every willing to prepare the microorganisms of dadih.

**Figure 1. Flow Chart CW and Tapioca Processing**
4. Conclusions

Due to fermentation, there was a decrease in pH, initial pH 4.8 to 3.4 and 4.1. Protein was increased significantly from 2.28 (control) into 7.48 (isolate dadih) and 4.94 (yeast dadih). Fat improved from 0.68 (control) to 2.64 (isolate dadih) and 1.26 (yeast dadih). CW could be fermented by isolat dadih or yeast/yeast dadih while dominant microorganism in dadih was Lactobacillus plantarum. Conventional sources of protein were very expensive, this was why, efforts to find nonconventional sources were of much concern. By using microorganism from dadih, livestockers could fermented CW even if they live in the very remote area without need to buy commercial bioactivator.

Recommendation

It is necessary to test the fermented CW in the sack by using a dadih starter with an incubation period of 1 month and 2 months with the condition of sack lies at the location of the sale. In vivo research is worth trying for cow, sheep and pig.

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