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Research Article

Selection of Local Microorganism Solutions as Crude Fiber Digester in *Areca catechu* L. Peel Waste for Producing Poultry Feed Additives

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Abstract

Background and Objective: *Areca catechu* L. is an herbal plant that contains polyphenol (flavonoids and tannin) and alkaloid (arecholine, arecolidine, guvacoline, guvacine and isoguvacine) compounds. Both polyphenols and alkaloids are known to be anthelmintic as well as antibacterial, antifungal, anti-inflammatory, antioxidant and cholesterol lowering compounds for poultry and dogs. When *Areca catechu* L. seeds are harvested, they produce 76% *Areca catechu* L. peel waste, which can be potentially used as a poultry feed additive to increase poultry performance. *Areca catechu* L. peel waste contains as much as 1.693% total polyphenols, 1.383% total alkaloids, 1.466% catechins and 456.59 mg/100 g tannins. The utilization of *Areca catechu* L. peel waste as a poultry feed additive is limited due to the high crude fiber content, which can be as high as 47.02%; the poultry digestive tract does not produce cellulase to hydrolyze crude fiber. The fermentation of *Areca catechu* peel waste with a local microorganism solution that produces cellulase is one solution for lowering crude fiber in *Areca catechu* L. peel waste. This experiment was performed to select the best local microorganism solution from different sources as a crude fiber digester in *Areca catechu* peel waste to produce poultry feed additives. **Materials and Methods**: This experiment was performed with a completely randomized design using five different sources of local microorganism solutions derived from bamboo sprouts, banana corms, mixed fruit waste, mixed vegetable waste and rice waste. As much as 500 mL of each local microorganism solution was used to ferment 200 g of *Areca catechu* L. peel waste for 2 weeks and each treatment was repeated four times. Crude fiber and dry matter were the parameters in this experiment and the total number of colonies of bacteria and fungi was calculated before and after fermentation with each local microorganism solution. **Results:** The fermentation of *Areca catechu* L. peel waste with different sources of local microorganism solutions significantly lowered crude fiber and dry matter ($p<0.05$) and the total number of colonies of bacteria and fungi decreased after the fermentation process, except for the colony of bacteria and fungi in the local microorganism solution from bamboo sprouts, which increased. **Conclusion:** Mixed vegetable waste was the best source of local microorganism solutions for lowering crude fiber in *Areca catechu* L. peel waste and the crude fiber decreased from 47.02-25.95%.

Key words: Alkaloids, *Areca catechu* L. peel waste, crude fiber, local microorganism solution, mixed vegetable waste, polyphenol

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Feed ingredients are generally obtained from agroindustrial materials because they do not compete with human food. Previous research on using and processing agro industrial wastes has been conducted for poultry feed ingredients. Tomato waste can be used up to 7% in broiler rations[1] and 12% in laying hen rations[2]. Nuraini *et al.*[3] also reported that fermentation using *Phanerochaete chrysosporium* and *Neurospora crassa* can improve the quality of durian fruit waste for poultry feed. The other agro industrial waste that has the potential to be used as feed additives for poultry is *Areca catechu* L.

Areca catechu L. is well known as an herbal plant that contains several active compounds, such as polyphenols including flavonoids and tannins[4] and it contains as much as 15% of these compounds. *Areca catechu* L. also contains alkaloids including arecholine, arecolidine, guvacoline, guvacine and isoguvacine, which are as much as 0.3-0.6% of the total composition. These active compounds play roles as anti-inflammatory, antibacterial, antifungal, or antioxidant agents in poultry and dogs and Byun *et al.*[5] reported that they inhibited cholesterol absorption in mice, reducing glucose in the blood plasma. When betel nuts are harvested, the process produces 76% fresh *Areca catechu* peel waste that has not been widely processed and utilized by the wider community; thus, it becomes decaying waste. Proximate analysis of *Areca catechu* peel waste demonstrated 65.41% water, 34.59% dry matter, 2.22% protein, 0.15% fat, 47.02% crude fiber, 0.28% Ca, 0.36% P and 2,495 kcal kg^{-1} [6] of metabolic energy, while Van Soest et al.[7] analysis showed that *Areca catechu* peel waste contained 59.07% NDF, 44.74% ADF, 27.44% cellulose, 14.32% hemicellulose and 17.30% lignin[6]. *Areca catechu* peel waste is also reported to contain active compounds of tannin polyphenols. In the Yaki, *Areca catechu* peel (*Areca vestiaria Giseke*) was found to have 3.16 mg kg^{-1} of total phenolic compounds with antioxidant activity of 54.11%[8]. *Areca catechu* peel extract was reported as an anti-microbial agent against *C. albicans* in humans[9]. Based on the content of nutrients and active compounds found in *Areca catechu* peel waste, it is possible to use it as a feed additive to improve the health and performance of poultry but the high content of crude fiber and lignin limits its use in poultry rations. The high crude fiber content of *Areca catechu* peel waste can be overcome with fermentation using a local microorganism solution that is cellulolytic and lignolytic. Local microorganism solution is a solution of bacteria, fungi or mold that is naturally present in an ingredient and it can be obtained from bamboo sprouts, banana corms, mixed fruit waste, mixed vegetable waste and rice waste[10]. Local microorganisms that contain cellulolytic and lignolytic enzymes potentially degrade organic matter that decomposes to produce compost, organic fertilizer and fungicides[11]. The results of processing *Areca catechu* peel waste with a local microorganism solution to overcome the high crude fiber and lignin contents through the fermentation method for use as a feed additive in poultry feeds have not yet been reported. Therefore, this study was conducted to evaluate the best local microorganisms from different sources (bamboo sprouts, banana corms, mixed fruit waste, mixed vegetable waste and rice waste) to lower the crude fiber content of *Areca catechu* peel waste.

MATERIALS AND METHODS

***Areca catechu* L. peel waste:** *Areca catechu* L. peel waste was collected from harvested *Areca catechu* L. seeds at a local farm field in West Sumatera Province, Indonesia.

Preparing the local microorganism solution from rice waste: The local microorganisms from rice waste were prepared by mixing 500 g rice waste with 1 L fresh water and 70 g sugar in a plastic bowl and then the mixture was fermented for 1 week.

Preparing the local microorganism solution from bamboo sprouts: The bamboo sprouts were harvested from a local farm field and 1000 sprouts were chopped into cubes and placed in a plastic bowl. A total of 3 L of water waste from washing the rice and 200 g of palm sugar were added to the bowl and then the mixtures were fermented for 15 days. Fermentation was stopped by centrifugal separation of the solution. The resulting solution was used for *Areca catechu* L. peel fermentation.

Preparing the local microorganism solution from banana corms: The Banana corms were harvested from a local banana farm field in West Sumatera Province, Indonesia. The banana corm (1000 g) was chopped into cubes in combination with 2 L of water from washing the rice and 200 g of brown sugar was mixed and placed in a plastic bowl and fermented for 15 days.

Preparing the local microorganism solution from mixed fruit waste: Avocado, banana, papaya, pineapple, watermelon, mango and guava (2 kg of each fruit) were

chopped into cubes and mixed in a plastic bowl. Then, 2 L of coconut water and 200 g of brown sugar were added to the fruit mixture and it fermented for 15 days. Fermentation was stopped by separating the solution with centrifugation.

Preparing the local microorganism solution from mixed vegetable waste: Radish, horseradish, mustard, kale and spinach waste (10 kg of each) were chopped and placed in a plastic bowl with 5% NaCl and 10 L of water from washing the rice; the mixture was fermented for 3 weeks. Fermentation was stopped by separation of the solution with centrifugation and then two percent brown sugar was added to the solution.

Experimental design: The experiment was performed in a completely randomized design with different local microorganism solutions (rice waste local microorganism solution, bamboo sprout local microorganism solution, banana corm local microorganism solution, mixed fruit waste local microorganism solution and mixed vegetable waste local microorganism solution) for fermentation of the *Areca catechu* L. peel waste and each treatment was replicated four times.

Parameters: Crude fiber and dry matter were measured in this experiment and the number of colonies of bacteria and fungi after fermentation with each local microorganism solution was calculated.

Fermentation of *Areca catechu* L. peel waste with the different local microorganism solutions: For the treatment, 200 g of fresh *Areca catechu* L. peel waste powder was placed into different plastic bowls and 500 mL of the different local microorganism solutions (from rice waste, bamboo sprouts, banana corms, mixed fruit waste and mixed vegetable waste) was poured into each plastic bowl. All of the fresh *Areca catechu* L. peel waste powder in the plastic bowls was fermented for two weeks. Fermentation was stopped by drying the fermentation product in an oven at 60°C for 2-3 days.

Statistical analysis: The data were statistically analyzed by one-way ANOVA. The differences among treatments were determined using Duncan's multiple range test[12]. If the treatments were significant, the data were analyzed using Duncan's multiple range test at 5% level of significance.

RESULTS

Crude fiber: The different local microorganism solutions significantly decreased the crude fiber content of the *Areca catechu* L. peel waste ($p<0.05$) after fermentation. The data are shown in Table 1.

Dry matter: After fermentation, the dry matter content of the *Areca catechu* L. peel waste was affected by the different local microorganism solutions ($p<0.05$). The data are shown in Table 2.

The total number of colonies of bacteria and fungi are shown in Table 3.

Table 1: Average crude fiber in the *Areca catechu* L. peel waste powder after fermentation with different local microorganism solutions (%)

Local microorganism solution	Crude fiber
Mixed vegetable waste	25.95^{a}
Mixed fruit waste	31.91^{ab}
Bamboo Sprout	40.13^{c}
Banana corm	31.51^{ab}
Rice waste	36.30^{bc}

Table 2: Average dry matter of the *areca catechu* L. peel waste powder after fermentation by different types of local microorganism solutions for 2 weeks, on a dry weight basis (%)

Local microorganism solution	Dry matter
Mixed vegetable waste	12.27^{b}
Mixed fruit waste	16.61^{ab}
Bamboo sprout	27.80^{a}
Banana corm	11.38^{b}
Rice waste	9.86^{b}

Table 3: Average total number of colonies of bacteria and fungi in each local microorganism solution type before and after fermentation of *Areca catechu* L. peel waste for 2 weeks

Type of local microorganism solution	Total No. of colonies of bacteria and fungi before fermented of *Areca catechu* L. peel waste (CFU)		Total No. of colonies of bacteria and after fermented *Areca catechu* L. peel waste for two weeks (CFU)	
	Bacteria	Fungi	Bacteria	Fungi
Mixed vegetable waste	123×10^{8}	225×10^{8}	68.5×10^{8}	96.25×10^{8}
Mixed fruit waste	195×10^{8}	154×10^{8}	7.5×10^{5}	10.5×10^{5}
Bamboo sprout	480×10^{8}	280×10^{8}	9.75×10^{10}	11.5×10^{10}
Banana corm	193×10^{8}	138×10^{8}	6.25×10^{5}	10.25×10^{5}
Rice waste	136×10^{8}	133×10^{8}	260.25×10^{6}	216.5×10^{6}

DISCUSSION

The average crude fiber content found in the *Areca catechu* L. peel waste decreased after fermentation with the different local microorganism solutions compared to the crude fiber content before fermentation. Mahata *et al.*[6] stated that the crude fiber content found in unprocessed *Areca catechu* L. peel waste was 47.02%. In this study, the crude fiber content of the *Areca catechu* L. peel waste after being fermented with the local microorganism solution from mixed vegetable waste was 25.95%, the local microorganism solution from banana corm was 31.51%, the local microorganism solution from mixed fruit waste was 31.91%, the local microorganism solution from rice waste was 36.30% and the local microorganism from bamboo sprout was 40.13%. In this experiment, the capability of the microbial solutions from mixed vegetable waste, mixed fruit waste and banana corm to degrade the crude fiber in *Areca catechu* L. peel waste was equivalent but the lowest amount of crude fiber was found in the *Areca catechu* L. peel waste fermented by the mixed vegetable waste (25.95%). It is predicted that microbes found in mixed vegetable waste are more active in degrading crude fiber. Adrizal *et al.*[10] stated that the ability of local microorganism solutions from mixed vegetable waste to degrade crude fiber found in pineapple waste was equivalent to the ability of local microorganism solutions from bamboo sprouts with fermentation durations of 1 and 2 weeks. Furthermore, the depletion of *Areca catechu* L. peel waste dry matter fermented with the local microorganism solution from mixed vegetable waste, mixed fruit waste, banana corm waste and rice waste was equivalent but different from that with the local microorganism solution from bamboo sprouts. Lindung[13] reported that there are several types of microorganisms in local microorganism solutions from bamboo sprouts, such as *Rhizobium* sp., *Azospirillum* sp., *Azotobacter* sp., *Pseudomonas* sp., *Bacillus* sp. and phosphate bacteria. The lowest dry matter was found in the *Areca catechu* L. peel waste that was fermented with the local microorganism solution from rice waste (9.86%). The local microorganism solution from rice waste is dominated by *Neurospora* sp. Sastraatmadja[14] stated that starter *Neurospora* sp. mold grew well in rice powder for the fermented food called oncom in Indonesia. This mold can produce cellulase enzymes[15] and several other enzymes, such as proteases[16] and lipases[17], which were the enzymes produced by the fungus that degraded the dry matter of the *Areca catechu* L. peel waste during the fermentation process, unlike those from the microbes found in the other local microorganism solutions. The mean number of colonies of bacteria and fungi found in all the local microorganism solutions in this study decreased after the *Areca catechu* L. peel waste was fermented for 2 weeks. This result was related to the amount of nutrients that also decreased to meet the needs of the microbes and to the microbes entering the death phase of their life cycle. However, the total number of microbial colonies (bacteria and fungi) in the local microorganism solution from bamboo sprout waste increased after the *Areca catechu* L. peel waste was fermented for 2 weeks. The increase is due to the microbes in the local microorganism solution from bamboo sprouts not entering the death phase of their life cycle; thus, the total number of colonies was greater than that from the other local microorganism solutions.

CONCLUSION

Mixed vegetable waste was the best local microorganism solution for lowering crude fiber in *Areca catechu* L. peel waste and the crude fiber decreased from 47.02-25.95%.

SIGNIFICANCE STATEMENT

This study discovered a method for lowering crude fiber in *Areca catechu* L. peel waste via fermentation with a local microorganism solution from mixed vegetable waste. This method could be beneficial in improving the digestibility of nutrients, especially catechin compounds, from *Areca catechu* L. peel waste used as a feed additive in poultry rations. The method in this study could be easily adopted by farmers and the material to produce the local microorganism solution from mixed vegetable waste is cheap. There have been no other reports from other researchers about the utilization of local microorganism solutions from mixed vegetable waste to ferment *Areca catechu* L. peel waste. Thus, a new method for lowering crude fiber in *Areca catechu* L. peel waste was found.

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