Effect of Nutritional Supplements of Fermented and Unfermented Betel Nutshell Waste in Performance of Broiler Chickens

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ABSTRACT

An experiment was conducted to see the broiler’s performance fed with fermented and unfermented betel nutshell. Harvesting betel nut seed leaved peel as a waste that contains some nutrients and active ingredients. This waste was useful for poultry feed, to maintain poultry performance and to reduce cholesterol. The aim of this research was to fermented betel nutshell waste with indigenous microorganism from a vegetable waste mixture that produced cellulase to reduce their crude fiber before feeding to the broiler. The nutrient profile and metabolizable energy of fermented betel nutshell in dry matter based showed 15.96 % water, 84.04 % dry matter, 23.69 % crude fiber, 10.39 % crude protein, 0.31 % crude fat, 2.60% calcium, 1.26 % phosphorus, and 1360.44 kcal/kg. In this experiment betel nutshell waste from a local farmer in Indonesia, and 80 birds from the broiler strain CP 707 from Charoen Pockphand were used. The experiment was conducted in a completely randomized design with different levels of fermented betel nutshell waste (0, 2, 4, and 6%) and 6 % of unfermented of betel nutshell waste in the broiler ration. Each treatment was repeated four times. The measured values included daily feed intake, daily weight gain, feed conversion, body weight, carcass content, abdominal fat pads, lipoprotein profile (total cholesterol, low-density lipoprotein (LDL), and high-density lipoprotein (HDL) in serum of broiler chickens. The results indicated that fermented betel nutshell waste significantly increased daily feed intake, daily body weight gain, carcass content, and decreased abdominal fat pad, total cholesterol, HDL, and LDL of broiler chickens, however did not affect the feed conversion. The fermentation of betel nutshell with indigenous microorganism from vegetable waste mixture could be used up to 6 % in the broiler ration and had a positive effect on performance, blood serum lipoprotein, and carcass quality of broiler chickens.

Key words: Betel nutshell waste, Broiler, Crude fiber, Fermented betel nutshell waste, Performance

INTRODUCTION

The broiler is one of the potential livestock to meet the demand for animal protein in Indonesian society. The consumption of chicken meat increases from year to year. According to Livestock and Animal Health Statistics (2018), broiler meat production on 2016 was 1.905.497 tons, on 2017 it was 2.046.794 tons and on 2018 it was even 2.144.013 tons. The high consumption of broiler meat in Indonesia due to the price of broiler meat was cheaper than local chicken meat and beef. The poultry industry needed food availability on a continuous basis.

Indonesia still imports some poultry feeds such as corn, soybean meal, and fish flour. According to ID Indonesia (2015), Indonesia was the country in the Southeast Asia region that bought soybean meal in the largest amount, in the 2013/2014 period, the top four soybean meal importers in Southeast Asia were Indonesia (4 million tons), Vietnam (3.3 million tons), Thailand (2.7 million tons), and Malaysia (1.6 million tons). On 2017 corn was imported from Indonesia at 360.355 tons, and it reached to 481.471 tons on 2018 (Situmorang, 2018). Import feed increased feed costs in the poultry industry, so Indonesia should look for alternative feeds to increase the availability of poultry feed and reduce imported feed. In addition to the diet that could reduce fat and cholesterol in broiler meat, which was important to the mixture in broiler diet, as consumers with limited fat intake were considering consuming broiler meat, the increasing growth rate of broilers followed by increasing fat deposition (Tumova and Teimouri, 2010). Furthermore, excess fat in poultry was one of the main problems faced by the poultry industry (Zhou et al., 2006).

Agriculture wastes could be used as unconventional poultry feeds, and it still contains active compounds like catechin, anthocyanin, lycopene to reduce fat. Harvesting
of seed betel nut will produce betel nutshell waste yielded up to 76% of the weight of fresh betel nuts (Mahata et al., 2018). It is reported that areca nut production in Indonesia reached 1,100.35 tons in 2016 (Badan Pusat Statistik Indonesia, 2016). The high production of betel nut follows with high betel nutshell waste and is available throughout the year.

Proximate analysis and energy metabolism indicated that betel nut shell waste had contained 65.41% water, 34.59% dry matter, 2.22% crude protein, 0.15% crude fat, 47.02% crude fiber, 0.28% calcium, 0.36% phosphorus, and 2.495 kcal/kg energy metabolism besides that betel nutshell waste contained 1.466% catechin (Mahata et al., 2018). The high crude fiber in betel nutshell waste was an inhibiting factor to be used in poultry ration. The poultry’s digestive tract does not produce enzymes (cellulose, hemicellulose, and lignocellulose) to hydrolize crude fiber, therefore the utilization of betel nutshell in poultry ration would be limited and needed processing before feeding to poultry. To overcome the high crude fiber content in betel nutshell waste, it could be fermented with Indigenous Microorganisms (IMO) which had the required enzymes to degrade the crude fiber. Adrizal et al. (2017) reported several IMO from the vegetable waste mixture, such as bamboo sprout, fruit waste mixture, rice water waste, banana corm to reduce the crude fiber. The best IMO was obtained from bamboo sprout to reduce the crude fiber in the pineapple (Ananas comosus L. Merr.) waste, also it was founded that the crude fiber in the pineapple waste decreased from 24% to 17.16% after fermentation. Furthermore, Mahata et al. (2018) stated that the obtained IMO from the vegetable waste mixture was the best from other IMO (bamboo sprout, fruit waste mix, rice water waste and banana corm) to degrade the crude fiber in the betel nutshell waste. Crude fiber and dry matter content of fermented betel nutshell waste with IMO from vegetable waste mixture was 12.27% dry matter and 25.95% crude fiber (Mahata et al., 2019). In addition, fermented betel nutshell waste contained some chemical active compounds and fatty acids such as 0.174% catechin, 35.420 mg/100g tannins, 0.663% laurate, 34.745% miristate, 55.116% palmitate, 0.527% stearate, 3.731% oleic, and 0.618% linoleate (Mahata et al., 2018).

Catechin contained in betel nutshell waste is a phytochemical substance that is naturally produced and contains flavonoid compounds. Catechin potentially improved carcass and meat quality of broiler through reducing cholesterol and fat. According to Kara et al. (2016a), catechins had effective antioxidant, hypoglycemic and hypocholesterolemic properties, and also potentially increased meat quality and had no negative effects on quail performance. Some researchers reported that the administration of catechins reduced total cholesterol, and it did not affect total protein levels in the blood serum of poultry (Abdo et al., 2010; Ariana et al., 2011; Kara et al., 2016b). Furthermore, Tang et al. (2002) indicated that the catechin could reduce fat in broiler meat. Also Yunarto et al. (2015) reported that the administration of extracted catechin with ethyl acetate from gambier (Uncaria gambir Roxb.) leaf would reduce cholesterol in the blood serum of rat by inhibiting HMG-CoA reductase activity to synthesis mevalonate from HMG-CoA in cells, so the synthesis of total cholesterol and LDL would decrease, while HDL would increase. There is no published study about the effect of fermented betel nutshell waste with obtained IMO from the vegetable waste mixture on the performance and blood profile of broiler chickens, and so the present research was decided to carry out the effectiveness.

MATERIALS AND METHODS

Experimental site
The experiment was performed at the Universitas Andalas farm, Limau Manis Campus, West Sumatra Province, Indonesia. The area is located at an altitude of ± 255 meters above the sea level and approximately 15 kilometer from Padang, the capital city of West Sumatra Province (Website Universitas Andalas, 2019).

Dietary treatment
The feed consumed in this experiment was formulated according to the standards for broiler feed. The feeds in the diet were corn, commercial feed (bravo 511), rice bran, palm oil, bone meal, Fermented Betel Nutshell Waste (FBNSW) and Unfermented Betel Nutshell Waste (UBNSW). The five treatment rations used in this study were formulated to contain approximately 20% crude protein, and 2900 kcal/kg metabolic energy. The nutrient percentage and metabolic energy (kcal/kg) contained in the experiment ration are indicated in table 1.

Experimental design and treatments
Completely Randomized Design (CRD) with five treatments of FBNSW and UBNSW (0, 3, 4, and 6% FBNSW, and 6% UBNSW) were used in this study. Each treatment was repeated four times. Twenty pens were used for the 80 day-old chickens, and four chickens in each pen were randomly assigned.
Rearing condition

The chicken cages, food and drinking equipment were cleaned and disinfected before the day-old chickens arrived. The lighting for each cage was 100-watts. Food and drinking water were given *ad libitum*. Day-old chickens (Charoen Pokphand 707, commercial broiler strains) were purchased from poultry shop in Padang City, West Sumatra Province, Indonesia.

Measured characteristics

Daily feed intake was measured by calculating the difference between delivered and denied food which were recorded for a trial period (28 days) in the morning. The average daily feed intake per bird was calculated by dividing the total amount of feed provided and refusals for a trial period (28 days). Afterwards the feed intake was divided for 28 days (trial period). Daily weight gain was calculated as the difference between the mean final and mean initial body weights divided by the number of experimental days (28 days). Feed conversion: It was determined by dividing the average daily feed intake with a mean daily weight gain. Body weight at the end of the study (28 days) was weighed. Twelve hours before weighing, the chickens were fasted. One bird was taken randomly from each pen for measuring the body weight. The percentage carcass fraction was measured at the end of the experiment, and one broiler was randomly selected from each replication. The broiler was slaughtered and weighed after 12 hours starvation. Exception of lung and kidney, the viscera, head, Shank, trachea, heart, liver, gizzard, and skin of slaughter broiler were eviscerated and expressed as carcass weight (gram). Furthermore, carcass percentage was calculated by dividing the carcass weight (gram) by the body weight (gram), and then multiplying by 100%. The abdominal fat pad percentage was obtained by comparing abdominal fat pad weight with body weight and then multiplying by 100%. Total cholesterol of broiler blood serum was calculated by Elitech group (2012) method. Up to 10 microliter (µl) of broiler blood serum was pipetted and then poured in the test tube, and 1000 µl cholesterol reagent was added to the test tube, further mixed and incubated for 10 minutes. Afterwards, it was calculated by a photometer. The High-Density Lipoprotein (HDL) in the broiler blood serum was calculated by using the Elitech group (2012) method. Broiler blood serum was pipetted up to 250 µl, then the reagent of HDL up to 500 µl added afterwards centrifuged for 10 minutes at the speed of 2500 rpm. Centrifuge results (supernatant) pipetted up to 100 µl was added to the cholesterol reagent up to 1000 µl mixed, and incubated for 10 minutes, afterwards, it was calculated by using a photometer. The Low-Density Lipoprotein (LDL) levels were determined by using Friedewald et al. (1972) formula. LDL = Total cholesterol – HDL – 1/5 Triglyceride.

Statistical Analysis

All data were analyzed by analysis of variance (ANOVA). Duncan’s multiple range tests were used to determine the differences between the treatment agents (Stell and Torrie, 1995).

Table 1. Food compositions, nutrient content, and metabolic energy of experimental ration in broiler chickens

| Feed compositions (%)  | FBNSW | 2% FBNSW | 4% FBNSW | 6% FBNSW | 6% UBNSW |
|------------------------|-------|----------|----------|----------|----------|
| Corn                   | 30.00 | 29.50    | 29.00    | 28.50    | 26.50    |
| Soybean meal           | 20.00 | 20.00    | 20.00    | 20.00    | 21.00    |
| Coco nut oil           | 2.00  | 2.25     | 2.50     | 2.75     | 3.00     |
| Rice bran              | 11.50 | 9.75     | 8.00     | 6.25     | 7.00     |
| Fermented betel nutshell waste  | 0.00  | 2.00     | 4.00     | 6.00     | 0.00     |
| Unfermented betel nutshell waste | 0.00  | 0.00     | 0.00     | 0.00     | 6.00     |
| Bone meal              | 1.50  | 1.50     | 1.50     | 1.50     | 1.50     |
| Bravo 511 (commercial feed) | 35.00 | 35.00    | 35.00    | 35.00    | 35.00    |
| Total                  | 100.00| 100.00   | 100.00   | 100.00   | 100.00   |

Nutrient content and energy metabolism of experiment ration

| Nutrient and energy metabolism of experiment ration  | FBNSW | 2% FBNSW | 4% FBNSW | 6% FBNSW | 6% UBNSW |
|---------------------------------------------------|-------|----------|----------|----------|----------|
| Crude protein (%)                                  | 20.72 | 20.72    | 20.71    | 20.71    | 20.55    |
| Crude fat (%)                                      | 6.47  | 6.60     | 6.74     | 6.87     | 6.74     |
| Crude fiber (%)                                    | 5.44  | 5.66     | 5.89     | 6.11     | 8.04     |
| Calcium (%)                                        | 0.90  | 0.95     | 0.99     | 1.05     | 0.91     |
| Phosphorus (available) (%)                         | 0.52  | 0.54     | 0.56     | 0.58     | 0.36     |
| Energy metabolism (kcal/kg)                        | 2.930.45 | 2.933.88 | 2.937.32 | 2.940.75 | 2.925.42 |
| Catechin (g/kg ration)                             | 0.00  | 0.03     | 0.06     | 0.10     | 0.88     |

FBNSW = Fermented Betel Nutshell Waste, UBNSW = Unfermented Betel Nutshell Waste. g = gram, kcal = kilo calorie, kg = kilogram.
RESULTS AND DISCUSSION

The inclusion of FBNSW and UBNSW in broiler ration influenced daily feed intake and daily weight gain significantly ($p < 0.05$), while feed conversion did not affect ($p > 0.05$) (Table 2). The daily feed intake and daily weight gain of broilers consumed 2, 4 and 6% of FBNSW in ratio greater than 0% FBNSW, and 6% UBNSW. These results indicated that the diets which contained 2, 4, and 6% FBNSW were more palatable by broiler compare with diets contained 0% FBNSW and 6% UBNSW. Increasing FBNSW content in broiler ration made the ration color a little darker than ration with 0% FBNSW and 6% UBNSW, while the color of the ration with 6% UBNSW resembled the color of ration with 0% FBNSW, so that the palatability of both rations (0% FBNSW and 6% UBNSW) were the same. The changing in ration color caused by increasing the FBNSW utilization in the ration did not affect the palatability of the broiler, although the ration’s color was slightly darker.

### Table 2. Average daily feed intake, daily weight gain, and feed conversion of broiler chicken fed fermented betel nutshell waste and unfermented betel nutshell waste for 4 weeks

| Treatments   | Daily feed intake (gram/bird/d) | Daily weight gain (gram/bird/d) | Feed conversion |
|--------------|---------------------------------|---------------------------------|-----------------|
| 0% FBNSW     | 81.32$^a$                       | 51.24$^b$                       | 1.59            |
| 2% FBNSW     | 85.83$^a$                       | 55.40$^b$                       | 1.55            |
| 4% FBNSW     | 85.83$^a$                       | 54.86$^a$                       | 1.57            |
| 6% FBNSW     | 85.89$^a$                       | 54.34$^a$                       | 1.58            |
| 6% UBNSW     | 82.02$^b$                       | 49.53$^b$                       | 1.66            |
| SE           | 0.67                            | 0.73                            | 0.001           |

SE = Standard error, FBNSW = Fermented Betel Nutshell Waste, UBNSW = Unfermented Betel Nutshell Waste. $^a$, $^b$, $^c$ = Means in a row that are not followed by the same letters are significantly different at ($p < 0.05$). g = gram, d = day

According to Sulasmni et al. (2013), the ration’s colors with brownish-yellow was more palatable than ration with dark color. The reduction in daily feed intake of broiler on ration contained 6% UBNSW as the crude fiber content was high compared to ration contained FBNSW. Fermentation treatment will increase the various of nutrients digestibility such as organic matter, nitrogen, amino acids, fiber, and calcium, and also increase feedstuff palatability (Canibe and Jensen, 2012; Shahowna et al., 2013). In this experiment, it was found that the average range of daily feed intake of broiler (4-weeks-old) was 81.31 to 85.89 g/bird/d. These results were almost the same with the experiment reported by Selle et al. (2019), where the average of broiler’s daily feed intake of broiler chickens fed 28-days with sorghum for was between 83.36 and 89.25 (g/bird/d). The daily weight gain of the broiler specified according to feed consumption. A ration with 2, 4, and 6% FBNSW had an effect on increasing daily weight gain of the broiler and was above 0% FBNSW and 6% UBNSW. According to Uzer et al. (2013) the increase in body weight gain was closely related to the feed consumption. A ration with 6% UBNSW reduced the daily weight gain of the broiler as it contained a high proportion of crude fiber compared to other rations. According to Kras et al. (2013), high crude fiber in poultry rations was not recommended as it had a negative effects on nutrient utilization, low body weight gain, and bad conversion rate. In addition, due to the low daily weight gain of the broiler chickens, the present study was suspected to contain 6% UBNSW with high tannin contain. According to Mahata et al. (2018), the tannin content in UBNSW was 456.59 mg/100g, while in FBNSW was 354.52 mg/100g. Tannins inhibited the process of protein digestion, while protein is a substance needed for growth and muscle building. Cook (2000), indicated that tannin bound to proteins and reduced metabolic protein, disrupting growth. The average daily weight gain of a 4-week-old broiler in this study ranged from 49.53 to 55.40 g/bird/d. The results of this study were almost the same as those reported by Selle et al. (2019), where the average daily weight gain of broiler fed sorghum for 28 days was between 54.25 and 55.56 g/bird/d. Increasing daily feed intake and daily weight gain of broiler which consumed 2, 4 and 6% of FBNSW in the ration did not match the feed conversion, and the ration efficiency for all treatments (0, 2, 4, 6% FBNSW and 6% UBNSW) were the same, because the nutritional substances in all of the treatments for the normal life of the broiler is sufficient. In this experiment, protein and energy were adjusted to iso-protein and iso-energy for all rations. The feed conversion index increased when the ratio between the amount of energy in the ration and protein content was technically adjusted (Mookiah et al., 2014). Furthermore, Andriyanto et al. (2015) stated, the only factor that affected the value of feed conversion was the nutrition quality. The average feed conversion of a 4-week-old broilers in this study ranged from 1.55 to 1.66. The results of the present study were almost the same as those reported by Zampiga et al. (2018), where the average feed conversion of broiler feed conversion was between 1.494 and 1.524 for 33 days.

The inclusion of FBNSW and UBNSW in the broiler ration significantly affected body weight, percentage of abdominal fat pads, and percentage of carcass ($p > 0.05$) (Table 3). The body weights of the broilers, that consumed
rations with 2, 4, and 6% of FBNSW were higher than the body weight of broilers, consumed 0% FBNSW and 6% UFBNWSW. The high body weight of the broiler, which consumed 2, 4, and 6% of FBNSW, was associated with high daily feed intake and daily weight gain in these treatments so that nutrient uptake to grow and produce high body weight was met. In addition, the digestibility of FBNSW in the ration was higher than the rations contained UBNSW in terms of fermentation efficiency, and the acceleration of digestive enzymes in the broiler digestive tract on FBNSW was optimum to better utilize nutrients from broilers for growth.

**Table 3.** Average body weight, abdominal fat pad percentage, and carcass percentage of broiler chicken fed fermented betel nutshell waste and unfermented betel nutshell waste for 4 weeks.

| Treatments | Body weight (gram/bird) | Abdominal fat pad percentage (%) | Carcass percentage (%) |
|------------|-------------------------|---------------------------------|------------------------|
| 0% FBNSW   | 1204.00a                | 1.58b                           | 70.39c                 |
| 2% FBNSW   | 1361.50b                | 1.38a                           | 71.90c                 |
| 4% FBNSW   | 1315.75bc              | 0.95a                           | 72.37c                 |
| 6% FBNSW   | 1248.75bc              | 0.93a                           | 70.64c                 |
| 6% UBNSW   | 1162.50bc              | 0.90abc                         | 69.28c                 |
| SE         | 24.23                   | 1.15                            | 0.31                   |

SE = Standard error, FBNSW = Fermented Betel Nutshell Waste, UBNSW = Unfermented Betel Nutshell Waste. Means in a row that are not followed by the same letters are significantly different at (p < 0.05).

**Table 4.** Average lipoprotein (total cholesterol, LDL and HDL) of broiler chicken fed fermented betel nutshell waste and unfermented betel nutshell waste for 4 weeks.

| Treatments | Total cholesterol (mg/dl) | Low-density lipoprotein (mg/dl) | High-density lipoprotein (mg/dl) |
|------------|---------------------------|---------------------------------|----------------------------------|
| 0% FBNSW   | 160.50ab                 | 120.25ab                        | 30.62ab                          |
| 2% FBNSW   | 149.50bc                 | 109.25bc                        | 30.55bc                          |
| 4% FBNSW   | 143.75bc                 | 105.75bc                        | 28.00bc                          |
| 6% FBNSW   | 125.25cd                 | 87.25cd                         | 29.00cd                          |
| 6% UBNSW   | 153.00bc                 | 109.25bc                        | 33.75c                           |
| SE         | 2.14                     | 1.97                            | 0.90                             |

SE = standard error, FBNSW = Fermented Betel Nutshell Waste, UBNSW = Unfermented Betel Nutshell Waste. Means in a row that are not followed by the same letters are significantly different at (p < 0.05).

According to Sari and Purwadaria (2004) generally, all fermented end-products typically contained compounds that are simpler and more digestible than the original ingredients. The low body weight of the broiler in treatment 6% UBNSW was caused by a higher tannin content (456.59 mg/100g) than the treatments 2, 4, and 6 in which FBNSW was 354.52 mg/100g (Mahata et al., 2018). Tannin could not be digested by poultry because the digestive tract of poultry did not produce tannase enzyme to hydrolyze tannins, and tannin bind the protein in UBNSW. In addition, tannin affected the reduction of feed consumption. Anita et al. (2012) reported that the feed consumption of broiler chickens decreased up to 4.5% after feeding on aged tea leaves flour, as the tea leaves contained tannin the body weight of broiler chickens depended on the amount of protein consumed for meat production in its growth process. Although the body weight of the broiler in treatment 6% UBNSW was lower than treatment 2, 4, and 6% FBNSW, the body weight was the same as treatment 0% FBNSW in the ration.

Inclusion of FBNSW and UBNSW in ration reduced the abdominal fat pad percentage of the broiler, due to FBNSW and UBNSW contain catechin compounds. Catechin was a flavonoid class driven from polyphenol compound that could inhibit the formation and accumulation of fat by condensing catechin with bile salts as a fat solvent in the digestive tract of poultry. Therefore, the absorption process of fat in the digestive tract was disrupted and reduced, so that the production of abdominal fat was small. According to Koo and Cho (2004), catechin compounds contained in tea could reduce the fat content. The average percentage of broiler’s abdominal fat pad in this study was between 0.90 and 1.58%. The percentage of broiler carcasses in treatments 2 and 4% of FBNSW was higher than in the treatments 0 and 2% of FBNSW, and 6% of UBNSW (Table 3). The percentage of carcasses is closely related to body weight and percentage of abdominal fat pads. In the present study, the body-weight of broiler was also higher in 2, 4, and 6% FBNSW treatments than in the 0% FBNSW and 6% UBNSW treatments.

The inclusion of FBNSW and UBNSW in the broiler ration significantly affected total cholesterol, LDL, and HDL (p < 0.05) (Table 4). The total cholesterol in broiler blood serum was lower in the 6% FBNSW and 6% UBNSW treatments than in the 0, 2 and 4% FBNSW treatments in ration due to catechin content in FBNSW and UBNSW. According to Khalaji et al. (2011), revealed a significant reduction in total cholesterol in the broiler blood serum after feeding catechin containing green tea extract. The average total cholesterol level in blood serum was ranged from 125.25 to 160.50 mg/dL. The decreasing in total cholesterol content in broiler blood serum was also followed by a decrease in LDL due to the presence of tanase enzymes produced by IMO during the fermentation process.
process, and it reduced tannins in FBNSW. According to Velayutham et al. (2008), catechins reduce the production of apolipoprotein B, the main component of LDL, so that in this experiment, LDL decreased in broiler blood serum. As the total cholesterol decreased and affected the lower HDL synthesis in the liver of broiler chickens, the HDL levels in the blood serum of broiler chickens treated with 2, 4 and 6% also reduced.

CONCLUSION

The fermentation of betel nutshell waste by indigenous microorganism from vegetable waste mixture could be used up to 6 % in broiler ration which had a positive effect on performance, blood serum lipoprotein, and carcass quality of broiler chicken.

DECLARATION

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