Effect of the Substitution the Fish Meal with Shrimp Head Waste Fermented in Diet on Broiler Performance

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Abstract. The experiment aimed to investigate the effect of fish meal (FM) substitution with the shrimp head waste fermented (SHWF) in the diet on broiler performance. The SHWF is the shrimp waste fermented with Waretha inoculum containing Bacillus amyloliquefaciens bacteria. The materials used for this experiment were 100 day-old-chick (doc) Arbor Acres CP-707 strains unsexed. The experiment was arranged in Completely Randomized Design in five treatments and four replications with five chicks each. The broilers were randomly assigned in five different levels of SHWF as substitution of fish meal in the ration. The treatments were 0 % substitute (R0), 5 % FM substitute with SHWF (R1), 10 % FM substitute with SHWF (R2), 15 % FM substitute with SHWF (R3), and 20 % FM substitute with SHWF (R4) respectively. The result of this experiment indicated that feed consumption, weight gain, feed conversion ratio and body weight were not significantly (P > 0.05). However, the percentage of carcass and abdominal fat showed a significant effect (P <0.05). The conclusion of this experiment is that substitution FM with SHWF until 20% in broiler diet can maintain a performance broiler. In this treatment, were the consumption of 2428.28 grams/head, bodyweight gain 1330.07 g/head, feed conversion ratio of 1.82; body weight 1383.17 g, the percentage of carcasses was 73.86%, carcass fat was 2.67% and percentage of abdomen fat was 1.92 %.

1. Introduction
The high price of basic protein feeds raw material such as fish meal (FM), meat bone meal (MBM) and soybean meal greatly influences the profits gained by small-scale poultry farmers. This condition is caused by dependence on imported feed ingredients. At this time only yellow corn can be supplied domestically. On the order side, the corn and fish meal is the major source of energy and animal protein in the poultry ration. Efforts are needed to substitute the conventional ingredients with non-conventional feed ingredients, like a shrimp head waste (SHW).

The extent of by-products utilization as a feed ingredient depends on the cost of conventional feedstuff, safety to animal health and alternative use [1]. Therefore, the economics of crop residue and by-products utilization require intensive analytical investigation to formulate a least-cost ration for animal production. Some of the non-conventional feedstuffs use as a substitute for conventional feedstuff include cassava meal, flour dust, biscuit waste, bakery waste, sorghum sprout, cacao pod, poultry offal, hatchery waste, shrimp heads waste, plantain peels, etc. Shrimp head waste (SHW) is of special importance among the non-conventional feedstuffs in Indonesia. Fisheries and aquaculture products are important sources of healthy food and wealth worldwide. Market analysis indicates that the quantity and commercial value of crustaceans accounts for 9.7% and
22.4% of the total world fish production respectively [2]. Shrimp is the largest single product considering the commercial value, representing 15% of the value of all globally traded fishery products in 2013. Asia plays an important role in shrimp production, accounting for 85% of international shrimp aquaculture production and 74% of wild shrimp capture in 2013 [3].

As high-value seafood, shrimp is processed for meat, with the heads and hard carapace remaining as shrimp processing waste [4]. The industrial processing of shrimp produces massive quantities of solid waste that is a notable source of animal protein, chitin, carotenoids, and other bioactive compounds that are not appropriately utilized. Nevertheless, shrimp heads contain many valuable bioactive compounds, such as chitin, protein, calcium, and natural carotenoids [5], which may have important commercial applications in food, medicine, feed, cosmetics, aquaculture, biotechnology, etc. [6, 7]. Chitin is found in various kinds of organisms, such as crustaceans, fungi, insects, and algae.

The increased quantities of shrimp heads waste have raised environmental problems and are a waste of natural resources. Therefore, new sustainable technologies are required for more efficient recovery of the high-value products from shrimp heads waste. Traditionally, chemical treatments (strong alkali and acid) have been reported to recover chitin from the associated protein, minerals, lipids, and carotenoids in shrimp heads waste [8, 9]. This results in high-cost and harmful effluent wastewater. It is inappropriate for economic and environmental reasons as it damages the structure of the products [10, 11]. As an alternative process, enzymes used for the deproteinization of shrimp heads waste have been recently described and they avoid the hazards and pollution from the chemical techniques [12]. However, the application of commercial enzymes for industrial production at a large scale might be too costly. The exploitation of endogenous enzymes in the biomass can be a valid and affordable alternative. In fact, shrimp and fish heads are rich in endogenous enzymes (e.g., proteases, peptidase, and lipases) [13,14], that can be applied for autolysis and obtaining protein hydrolysate from fish and shrimp heads [15].

At the same time, fermentation has been described as an environmentally friendly, economical, flexible, and efficient technology in the utilization of biowaste. The fermentation of shrimp waste with protease or organic acid-producing bacteria promotes the production of a solid section of chitin and a liquor consisting of shrimp protein, minerals, and other components [16,17]. In previous studies, Streptococcus thermophilus [18], Bacillus licheniformis [19], Bacillus subtilis [20], and other bacteria have been applied for the deproteinization of crustacean waste to produce protein, antioxidant products, and chitin by liquid-state fermentation. In this research, the extraction of chitin and proteins from shrimp head by the combination of autolysis and fermentation was studied to meet the demands of higher productivity and less water consumption.

2. Materials and methods

2.1. Data collection and animal management

Probiotics from Waretha products containing Bacillus amyloliquefaciens inoculum were used for this study. The composition of the substrate and the dose of Waretha's inoculum in the fermentation process of shrimp waste to make shrimp head waste fermered products (SHWF) are show in Figure 1. The SHWF substrate consists of a mixture of 80% SHW and 20% rice bran and autoclave for 30 minutes, after which it is left at room temperature. After chilling, inoculated with 3% per kg of substrate using  the Waretha inukulum products and stirring evenly. And then incubated for 72 hours at 40 degrees C. After that, the SHWF product is dried in the sun. Finally, technical research in the form of a ration test to find out whether SHWF to replace local fish meal (FM) can be used in poultry rations.

The study to determine the effect of interaction between dose of inoculum Waretha (Bacillus amyloliquefaciens probiotic/BAP) and fermentation time using on nutrient content and quality of fermented shrimp waste flour were investigated. This research have show the best SHWF nutrient content obtained at treatment with 3% inoculum of Bacillus amyloliquefaciens probiotic (BAP) and 72 hours fermentation time with yield of 37.01% from 2 kg raw meterials, dry matter 81.78%, crude protein equal to 42.72%, crude fiber 11.01%, crude fat 2.41%, with nitrogen retention value equal to 66.60%, crude protein digestibility 72.07%, pigmen of asthaxantins 2.34 %, chitin contents 9.12 % and metabolizable energy 2585.72 kcal / kg [21].
2.2 Experimental birds and management

A total of One hundred day-old-chicks of broiler commercial Arbor Acres CP-707 strains were used for this experiment. The birds purchased from local poultry shop at Padang city and were selected from of a larger number after balancing for weight. The birds were individually weighed and use wing band and than randomly allotted to five treatment diet. There were 20 birds on each treatment and 5 birds per replicate in a completely randomized design. Experimental diets and drinking water were provided ad-libitum and study lasted 4 weeks. The water troughs were washed daily before fresh water was served. The broiler were environmental conditions. The birds were treated with broad-spectrum antibiotics and vaccinated before the experiment done. Antistress was administered occasionally during the experiment. Routine management practices were maintained.

The experiment diets composed of dry matter ingredients like yellow corn, rice bran, local fish meal, soybean meal, shrimp head waste fermented (SHWF), BR 511, coconut oil, and multivitamin mix are shown in Table 1. Five experiment diets, R0, R1, R2, R3, and R4 were formulated such that the diets were isonitrogenous 22.00 % and isocaloric 3000 kcal/kg. Diet R0 served as the control diet and containing 20 % fish meals and 0 % of shrimp head waste fermented. Diet R1, R2, R3, and R4 contained 5, 10, 15, and 20% of SHWF as substitute fish meals respectively. Other ingredients used
for ration formulation were obtained from a reputable feed mill. The composition of the experimental diets and calculated nutrient is shown in Table 1.

### Table 1. Composition of experimental broiler diets (as-fed basis)

| Ingredient          | Experimental diets |
|---------------------|--------------------|
|                     | R0 | R1 | R2 | R3 | R4 |
| Yellow corn         | 42.00 | 42.00 | 42.00 | 41.00 | 40.50 |
| Rice bran           | 5.00  | 5.00  | 5.00  | 5.50  | 5.50  |
| Fish meal           | 20.00 | 15.00 | 10.00 | 5.00  | -     |
| Soybean meal        | 27.50 | 26.50 | 26.00 | 26.00 | 26.00 |
| SHWF                | -   | 5.00  | 10.00 | 15.00 | 20.00 |
| BR 511a             | 4.50  | 5.50  | 6.00  | 6.00  | 6.50  |
| Coconut oil         | 0.50  | 0.50  | 0.50  | 1.00  | 1.00  |
| Top Mix (broiler)   | 0.50  | 0.50  | 0.50  | 0.50  | 0.50  |
| Total               | 100  | 100  | 100  | 100  | 100  |

Calculated analysis

|                   | R0 | R1 | R2 | R3 | R4 |
|-------------------|----|----|----|----|----|
| Crude protein (%) | 22.20 | 22.15 | 22.11 | 22.12 | 22.08 |
| Crude fat (%)     | 4.95  | 5.17  | 5.13  | 5.32  | 5.85  |
| Crude fiber (%)   | 3.82  | 4.68  | 4.70  | 5.04  | 5.14  |
| Calcium (%)       | 0.91  | 0.99  | 1.05  | 1.09  | 1.16  |
| Phosphorous (%)   | 0.49  | 0.48  | 0.45  | 0.52  | 0.51  |
| Lysine (%)        | 1.08  | 1.04  | 1.02  | 0.98  | 0.95  |
| Methionine (%)    | 0.52  | 0.49  | 0.49  | 0.47  | 0.45  |
| Metabolizable Energy (kcal/kg) | 3013.00 | 3008.96 | 3005.79 | 3008.91 | 3005.24 |

a = starter diets from PT. Charoen Pokphan Indonesia

2.3. Experimental design

The design of the experiment was Completely Randomized Design (CRD) with 5 treatments that were replicated 4 times and each replicated contained 5 birds. Data collected body weight, weight gain, feed consumption, feed conversion, carcass percentage, percentage abdomens fat and income over feed cost. Broiler performance data for feed consumption, weight gain, and feed conversion were collected for 4 weeks experimental period on weekly basis, while for percentage carcass and percentage fat abdomen were taken at the end of experimental. Feed conversion was calculated by dividing the feed consumption by the weight gain. Percentage carcass was calculated by dividing the carcass weight by body weight and multiple with 100 %, while percentage fat abdomen was taken by weight broiler abdomen fats and dividing with body weight and multiple with 100 %.

2.4. Chemical analysis

Proximate analysis and metabolizable energy of the test material and experimental diets were determined by the method of the Association of Official Analytical Chemists [23]. To compare the feed ingredients that were replaced with substitute feed ingredients, both ingredients the local fish meal and the shrimp head waste fermented were analyzed for contents and nutritional quality.

2.5. Statistical analysis

Data obtained in the study were subjected to analysis of variance (ANOVA) using Completely Randomised Design Significant means were further separated by the use of the Duncan Multiple Range Test [24].

3. Result and discussion

3.1. Proximate analysis of the SHWF products compared the local fish meals (FM)

Chemical composition of shrimp head waste fermented (SHWF) with bacteria of Bacillus amyloliquefaciens and local fish meals is shown in Table 2. SHWF product result fermented 3 days with 3% dose of the inoculum “Waretha” products (Bacillus amyloliquefaciens probiotic/BAP) had
the highest crude protein content after fermentation was 42.72%, nitrogen retention was 66.60%, and metabolizable energy was 2213 kcal/kg, and also have lowest of crude fiber and chitin content. The quality nutrient of SHWF almost the same or equivalent to the local fish meals.

Table 2. Proximate analysis of SHWF and local fish meals (as fed basis) and quality of nutrient

| Nutrient Components (%) | Feed ingredients | SHWF | Local fish meals |
|-------------------------|------------------|------|-----------------|
| Water                   | 11.43            | 10.10 |
| Crude protein           | 42.72            | 50.50 |
| Ash                     | 20.87            | 18.20 |
| Crude fiber             | 11.01            | 1.65  |
| Ether extract           | 2.41             | 6.40  |
| Calcium                 | 8.93             | 5.11  |
| Phosphorous             | 2.05             | 2.88  |
| Chitin                  | 9.12             | -     |
| Protein digestibility   | 72.07            | 90.0  |
| Metabolizable Energy    | 2213.00          | 2820  |
| Nitrogen retention      | 66.60            | -     |
| Methionine              | 1.44             | 1.63  |
| Lysine                  | 0.66             | 4.51  |
| Tryptophan              | 0.51             | 0.49  |

Source: Mirzah and Montesqrit [21]

3.2. Effect of substitute te local fish meal with the SHWF products on performance

Substitution the fish meal with shrimp head waste fermented (SHWF) by inoculum Waretha contains Bacillus amyloliquefaciens of bacteria. The performance of broiler in terms of feed consumption, weight gain, feed conversion, body weight, income over feed cost, carcass percentage, and percentage abdomens fat was investigated and the results are shown in Table 3. There were no significant differences in total feed consumption, weight gain, feed conversion ratio, and body weight (p > 0.05) among the five treatments. While the SHWF substitute is showed a significant difference (P < 0.05) on percentage carcass and percentage abdomen fat.

The treatment R4 had the highest feed consumption 2425.28 g for 4 weeks experimental period, while the least feed consumption was recorded in treatment R0 with an average 2444.81 g. Not effect significantly substitute local fish meals with the SHWF on feed consumption caused by the shrimp head waste fermentation process can increase palatability products and besides that, the nutritional content of the five experimental rations is the same. The experimental diets were isocaloric and isonitrogenous.

Table 3. Performance of broilers fed diets containing the level of SHWF as substitute the fish meal

| Parameter                  | R0       | R1       | R2       | R3       | R4       | SEM |
|----------------------------|----------|----------|----------|----------|----------|-----|
| Feed consumption (g)       | 2444.81  | 2442.94  | 2426.56  | 2436.57  | 2425.28  | 6.70 |
| Weight gain (g)            | 1332.38  | 1341.56  | 1330.50  | 1326.44  | 1330.07  | 5.37 |
| Feed conversion            | 1.83     | 1.82     | 1.82     | 1.84     | 1.82     | 0.01 |
| Body weight (g)            | 1391.31  | 1403.25  | 1390.87  | 1385.31  | 1383.17  | 7.93 |
| Carcass percentage (%)     | 67.17a   | 70.11a   | 69.07a   | 71.23a   | 73.86b   | 3.19 |
| Percentage abdomen fat (%) | 2.52a    | 2.18a    | 2.23a    | 2.16a    | 1.92b    | 0.38 |
| Income over feed cost (IDR) | 4955.00 | 4965.00 | 5570.00 | 6066.00 | 6660.00 |
|-----------------------------|---------|---------|---------|---------|---------|

\[ \text{a,b : values in the same row with different superscripts are significantly different (p < 0.05)} \]

The inclusion of 20% SHWF as a substitute for the local fish meal in the diet did not decrease the diet palatability because the color and flavor of diet not significantly different from the control diet (diet with 0% SHWF). According to [25], stated that color and flavor diet are factors affecting broiler feed consumption, the bright color more palatable than dark color. The enhancement until 20% SHWF as a substitute for the local fish meal in the diet is not decreased palatability of the treatment diets and to obtain the same feed consumption between treatment diets. Previously, [26], reported that fermented products can produce flavors that are preferred by broiler and have also several vitamins (B1, B2, B12) that are preferred compared to the original raw materials.

Probiotics play an important role in livestock growth. [27], the fermentation process using \( Bacillus \) \textit{amyloliquefaciens} probiotic (BAP) can provide beneficial physical and chemical changes such as flavor, palatability, texture, and digestibility better than the original ingredients. The enzymes produced by microorganisms can degradation complex compounds such as carbohydrates, proteins, and fats into simpler compounds such as glucose, amino acids, and fatty acids. This proves that the cellulase enzyme produced by \( Bacillus \) \textit{amyloliquefaciens} can degradation some cellulose in the shrimp head waste. Additionally, [28, 29, 30], that processing shrimp head waste using chitinase producing bacteria will increase the digestibility and solubility crude proteins of feed.

The results of those studies showed that there were no significant effects on feed consumption. So that this condition will result in weight gain, feed conversion ratio, and body weight will be the same. The SHWF given in the formulation of the diet can be functional feeds and helps in the digestion process and the development of the gastrointestinal tract as well as synthesis and secretion of digestive enzymes determine the further conversion and utilization of nutrients [31]. However, the SHWF products can be probiotics in the treatment diet and also interact with the host by producing lactic acid and enzymes, then the bird not only has better intestinal health but can also more easily digest the feed, which improves growth performance. Probiotics composed of spores of \( Bacillus \) \textit{amyloliquefaciens} are known to produce α-amylase, which is used in hydrolysis of the starch [32, 33]. This strain also produces a large number of extracellular enzymes such as cellulase [34] and proteases [35], which can improve the digestion of nutrients. Additionally, [36] reported that the improved protein, fat, and starch digestibility in broilers fed diets supplemented with \( Bacillus \) \textit{amyloliquefaciens} products (106 CFU/g of feed). In present study, the same growth performance of R4 treatment diets in 20% the SHWF of broiler compared with the control diets, is believed to have been induced by the total effects of probiotics of SHWF action, including maintenance of normal intestinal microflora and increased digestive enzymes secretion that improved nutrient digestibility and decreased ammonia production. Additionally, the same growth performance, especially on weight gain, feed conversion ratio, and body weight caused all treatment diets to have the same nutrient contents and quality.

The high nutrient quality of SHWF could also affect body weight and FCR. Previously, [22], reported that fermentation of shrimp head waste with inoculum Waretha (\( Bacillus \) \textit{amyloliquefaciens}) was increased nutrient and quality of SHWF products, especially the crude protein, nitrogen retention, and bioavailability of energy. In a meta-analysis, [37], also reported a 1.7% increase in final body weight and 3.0% reduction in FCR in broiler fed diet supplemented with \( Bacillus \) \textit{amyloliquefaciens} products. And then [32], reported that improved growth performance in broiler fed a diet supplemented with α-amylase obtained from \( Bacillus \) \textit{amyloliquefaciens}. Thus, it is evident that, SHWF as functional fed in diet until 20% could improve equal with the growth performance of broiler control diets.

The percentage of carcass and abdomens fat were significantly different (\( P < 0.05 \)) in the five treatments diet. The highest carcass percentage and the lowest abdomens fat percentage deposition were obtained with R4 treatment diets (Table 3). Carcass percentage of R4 was significantly (\( P < 0.05 \)) higher than R0, R1, R2, and R4 treatments, it means protein quality in the SHWF can supply requirements of bird and number of spores Waretha products is 1022 CFU/g [22]. Probiotics play an important role in stabilizing the intestinal ecosystem of animals by enhancing the growth of beneficial bacteria and competing with pathogenic bacteria in the intestine [38, 39]. Lowest the abdomens fat
percentages deposition responses obtained with R4 treatment diets (100% local fish meal replacement by the 20% SHWF in diet). While abdomens fat percentages R4 were significantly (P < 0.05) lower than R0, R1, R2, and R3 diets, it means glucosamine from chitin degradation affects the amount of fat absorption and synthesis of fatty acid, so deposition of abdomens fat to be decreased. This research found, higher with [40], was found average abdomens fat percentage about 1.83%.

Results of income over feed cost (IOFC) were significant among the five treatments (Table 3). In practice, feed conversion ratio (FCR) can be calculated as the total feed intake (kg) divided by the total body weight at the end of production or experiment (kg) [41]. The value of FCR can measure the IOFC of diet in IDR. The highest IOFC of 6,660 IDR was recorded in treatment R4 (20% of SHWF was replacing 100% of local fish meal in diet) while the lowest IOFC 4,955 IDR was recorded in treatment R0 (control diet). Therefore, from the result of study, the IOFC improved with the increasing level of the SHWF in the experimental diets. The improvement in the IOFC as the level of the SHWF increased in diets indicated the presence’s novelty prices of the SHWF so that decreased prices of ration. In addition, these experiments, no mortality was recorded in all treatment. This implies that the SHWF has no toxic effects or any anti-nutritional factor in it since the SHWF is safe for animal consumption.

4. Conclusion
The conclusion of this experiment is that substitution local fish meals with the SHWF products until 20% in the broiler diet could be a maintenance performance broiler or substitute 100% local fish meal as an alternative of an animal protein source as much as 20% without any negative effect.

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