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Broiler Response to the Utilization of Fermented Palm Oil Sludge with *Phanerochaete chrysosporium* and *Neurospora crassa*

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ABSTRACT

Palm Oil Sludge (POS) had the potential as an ingredient of broiler ration. POS requires to be processed by fermentation of *P. chrysosporium* and *N. crassa* to increase nutritional value. The aim of this research was to evaluate the utilization of Fermented Palm Oil Sludge (FPOS) with *P. chrysosporium* and *N. crassa* in rations of broiler. One hundred and sixty mixed-sex a-day-old chicks (DOC) Cobb CP 707 breed of broiler procured from Charoen Pokphand Indonesia were used in this experiment. Broiler were placed in 20 wire cages; each unit consists of 8 birds. Feeding treatment was carried out for 35 days. Ration was composed of 22% iso-protein and 3000 kcal/kg of iso-metabolic energy. The design of this experiment is a completely randomized design with five treatments and four replications. Treatments were: T1) 0% FPOS (control diet), T2) 15% FPOS, T3) 20% FPOS, T4) 25% FPOS and T5) 30% FPOS in broiler ration. Feed intake, body weight gain, feed conversion ratio, body weight, carcass percentage, and abdominal fat percentage) were found to be significantly (P<0.05) decreased with any treatment except carcass weight (P>0.05) as compared to control group. In conclusion, FPOS with combination of *P. chrysosporium* and *N. crassa* was capable to be used up to 25% in rations of broiler.

Key words: Broiler, Fermented palm oil sludge, *Neurospora crassa*, *Phanerochaeta chrysosporium*.

INTRODUCTION

Palm Oil Sludge (POS) is waste product of crude palm oil production and makes up 2% of the yield from the fruit bunches but is often unused and considered a source of pollution. That is because it has a low nutritional value, being high in crude fiber and low in protein or amino acid content (Sinurat 2000). POS contains 13% crude protein, 12.3% crude fat, 32.1% crude fiber and 1105.9 kcal/kg metabolic energy (Mirnawati et al. 2017). Based on Noferdiman, 2004 research that palm oil sludge contains 7.3% hemicellulose and 14.2% lignin. Unlike its relatively high nutritional content, its utilization is still limited to 5% in poultry ration (Sinurat et al. 2000). That is due to the indigestibility of the crude fibers, especially cellulose and lignin.

In order to enhance the usage of POS in the broiler's diet, fermentation was conducted. Fermentation was used to increase digestibility by breaking down the fiber, while also enhancing flavor and aroma, finally resulted in the higher nutritional content of ingredients (Mirnawati et al. 2019a; Mirnawati et al. 2019b; Mirnawati et al. 2019c). High CF and lignin in POS required molds to produce enzymes to hydrolyze lignin i.e., *P. chrysosporium* mold. *P. chrysosporium* is white-rot mold which known for its capability to reduce lignin (Zeng et al. 2010), and produce high ligninase and cellulase (Howard et al. 2003). Noferdiman and Yani (2013) has shown that FPOS with 6% *P. chrysosporium* inoculums for eight days had 12.2% less crude fiber and 8,9% lignin. It also had 14.1% more crude protein and can replace standard chicken feed up to 15%.

Mirnawati et al. (2010) found that FPOS could increase the content of crude protein to 20.4%, crude fiber decreased to 23% and decreased crude fat to 3.7% and increase the digestibility of the crude fiber up to 48.4%, nitrogen retention 56.2% and metabolic energy 2024.3 kcal/kg. Based on results of Mirnawati et al. (2018), FPOS with *N. crassa* could not replace more than 22% of standard broiler ration. Mirnawati et al. (2019a) studied on FPOS of *P. chrysosporium* and *N. crassa* fungi, using a ratio of 4:

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1for 13 days. This gave: dry matter 89.3%, crude protein 26.2%, crude fiber14.5%, crude fat 2.2%, calcium 0.3%, phosphor 0.7%, lignin 14.5%, nitrogen retention 58.2%, a digestibility of crude fiber 57.7% and metabolic energy 2787.9 kcal/kg.

From the above data it may be seen that there is an increase in the content and quality of FPOS with *P. chrysosporium* and *N. crassa*. This result suggested that both *P. chrysosporium* and *N. crassa* used to fermented POS, would be a high-quality ingredient for poultry. This study aimed to specify the effect of feed ingredient on animal performance and determine the percentage in the ration. Field testing is required to search the optimal percentage of FPOS that could be utilized in broiler feed and its impact on performance.

MATERIALS AND METHODS

Animal Ethics

Animal experiment were carried out based on the Republic of Indonesia Law No. 18 of 2009 (section 66) chapter animal ethics that related to holding, rearing, and slaughtering and proper treatment and care for animals.

Broiler Maintenance

One hundred and sixty mixed-sex a-day-old breed cobb CP 707 broiler chicks from Charoen Pokphand Indonesia were raised in twenty 80x60x50 cm wire cages. Each cage held eight broiler and had its feeding and drinking stations and 60-watt incandescent lamps for heating and lighting. Rations were prepared by mixing commercial feed CP 511 with yellow corn, fish meal, soybean meal, rice bran, vegetable oil, top mix, and FPOS. Maintenance period was carried out up to five weeks (35 days).

Feeding Formulation

FPOS was made of 80% POS and 20% rice bran, which then fermented (at a room temperature) with *P. chrysosporium* and *N. crassa* with a ratio of 4:1 of each inoculum respectively. The product was incubated for 13 days in an incubator, got harvested, and then dried in an oven at temperature of 60ºC. After that, FPOS finally grounded and ready as feedstuff for poultry. The ration contained 22% iso-protein and 3000 kcal/kg of iso-metabolic energy. Details of composition were seen from Table 1.

Experimental Design

The research method used was an experimental method of Completely Randomized Design (CRD) with five treatments and four replications. Treatments were T1, T2, T3, T4, T5 with the use of 0, 15, 20, 25 and 30% fermented palm oil sludge (FPOS) respectively. The variables observed were feed intake (FI), body weight gain (BWG), feed conversion rate (FCR), body weight (BW), carcass weight (CW), carcass percentage (CP) and percentage of abdominal fat (AFP) for each broiler were measured following Maslami et al. (2019).

Statistical Analysis

The data collected were processed with ANOVA (Analysis of Variance) and DMRT (Duncan's Multiple Range Test) was performed as post hoc test (Steel and Torie 1995).

RESULTS

Feed Intake (FI)

Significant ($P<0.05$) effect was seen on analysis of statistic on FI after giving FPOS up to 30% in the ration. DMRT showed significant effect on Treatment T5 but non-significantly ($P>0.05$) different on other treatments (Fig. 1). Giving FPOS up to 25% in the ration can match the consumption of the control ration. However, feed consumption decreased in T5 treatment (30% FPOS).

Body Weight Gain (BWG)

Significant ($P<0.05$) effect also seen on statistical analysis of BWG after giving FPOS up to 30% in the ration. DMRT showed non-significantly different on treatment T1, T2, T3 and T4, but significant effect on Treatment T5 (Fig. 1). Giving FPOS up to 25% in the ration can match the body weight gain of the control ration. However, body weight gain decreased in T5 treatment (30% FPOS).

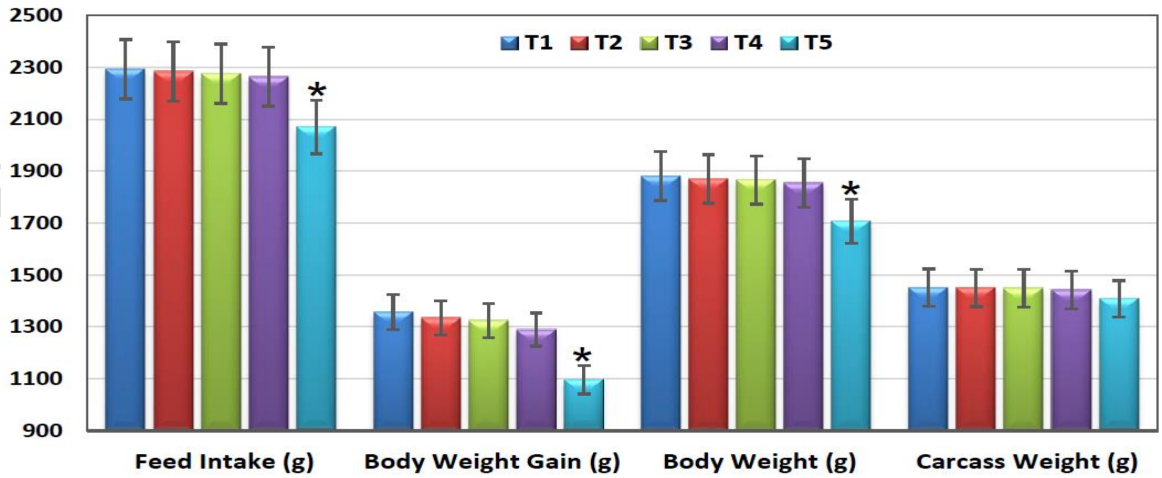

Fig. 1: Effect of dietary fermented palm oil sludge (FPOS) on broiler feed intake, body weight gain, body weight and carcass weight for 35 days. Inclusion of FPOS in T1 to T5 was 0% (T1-Control), 15% (T2), 20% (T3), 25% (T4), and 30% (T5), respectively. Bar bearing asterisk in a specific parameter differ significantly ($P<0.05$) than that of control group.

Table 1: Feeding composition (%) of treatment/rations

Feed ingredients	Treatment Ration				
	T1	T2	T3	T4	T5
CP 511 commercial feed	10	10	10	10	10
Yellow corn	53	45	42	40	39.5
Rice bran	6	4	4	3	1
Soybean meal	13	8	6	4	1.5
FPOS	0	15	20	25	30
Fish meal	16	16	16	16	16
Coconut oil	1.5	1.5	1.5	1.5	1.5
Top mix	0.5	0.5	0.5	0.5	0.5
Total	100	100	100	100	100

Table 2: Nutrient composition (%) and energy contents composition and metabolic energy (kcal/kg) in treatment feed

Item	Treatment Ration				
	T1	T2	T3	T4	T5
Crude protein	22.05	22.44	22.45	22.46	22.28
Crude fat	4.05	4.01	4.00	3.98	3.94
Crude fibers	4.03	5.46	5.97	6.42	6.79
Calcium	1.27	1.24	1.23	1.21	1.19
Phosphor	0.66	0.71	0.73	0.75	0.77
Energy metabolism	3028	3032	3025	3036	3039

Body Weight (BW)

Significant (P<0.05) effect was seen from analytical statistic of FBW after giving FPOS up to 30% in the ration. DMRT showed significant effect on Treatment T5 but non-significantly different on treatment T1, T2, T3 and T4 (Fig. 1). Giving FPOS up to 25% in the ration can match body weight of the control ration. However, body weight decreased in T5 treatment (30% FPOS).

Feed Conversion Ratio (FCR)

Statistical analysis of FCR showed significant effect (P<0.05) after giving FPOS up to 30% in the ration. DMRT showed non-significantly different on treatment T1, T2, T3 and T4, but significant effect on Treatment T5 (Fig. 2). Giving FPOS up to 25% in the ration can match the feed conversion of the control ration. However, body weight gain decreased in T5 treatment (30% FPOS).

Carcass Percentage (CP)

After administration of 30% FPOS in ration, analytical statistic on CP showed a significant effect (P<0.05). DMRT showed non-significantly different on treatment T1, T2, T3 and T4, but significant effect on T5 (Fig. 3). Giving FPOS up to 25% in the ration can match the carcass weight of the control ration. However, carcass weight decreased in T5 treatment (30% FPOS).

Abdominal Fat Percentage (AFP)

Statistical analysis on AFP (Fig. 4) showed a significant (P<0.05) effect after giving FPOS up to 30% compared to control treatment. However, giving FPOS up to 30% in the ration can't reach the AFP from control ration. Broiler abdominal fat percentage decreased in T2 treatment (15% FPOS).

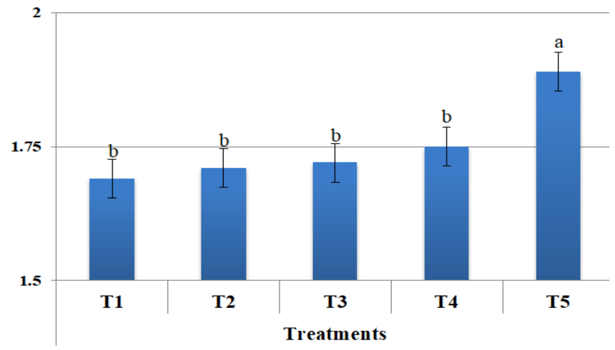

Fig. 2: Effect of dietary fermented palm oil sludge (FPOS) on broiler feed conversion ratio for 35 days. Inclusion of FPOS in T1 to T5 was 0, 15, 20, 25, and 30%, respectively. Bar bearing different letters differ significantly (P<0.05).

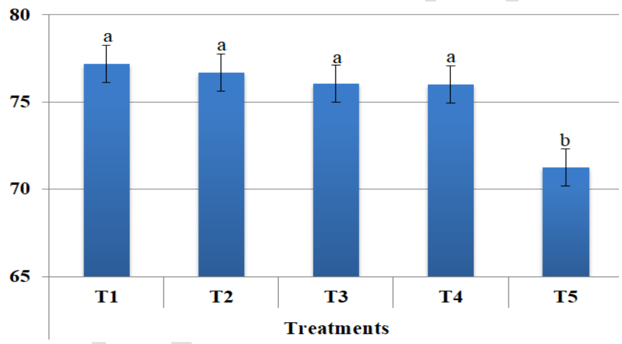

Fig. 3: Effect of dietary fermented palm oil sludge (FPOS) on broiler carcass percentage (%) for 35 days. Inclusion of FPOS in T1 to T5 was 0, 15, 20, 25, and 30%, respectively. Bar bearing different letters differ significantly (P<0.05).

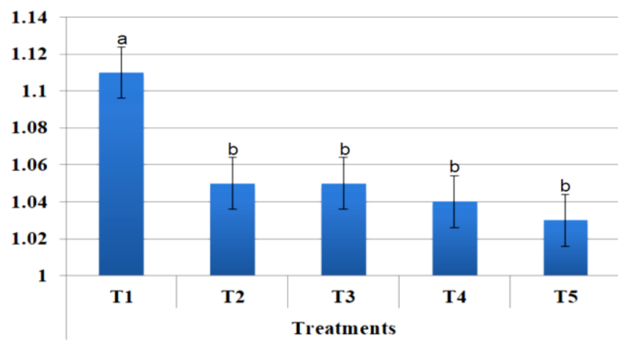

Fig. 4: Effect of dietary fermented palm oil sludge (FPOS) on broiler abdominal fat percentage (%) for 35 days. Inclusion of FPOS in T1 to T5 was 0, 15, 20, 25, and 30%, respectively. Bar bearing different letters differ significantly (P<0.05).

DISCUSSION

As fermentation occurred, FPOS product had good content and quality, because of the enzyme activity produced by microbes that can break down the complex into simple materials so that they are easier to digest by the livestock who consume them. Besides that, fermented products can increase aroma, taste and are more palatable and easy to digest, of course, will be affected to consumption (Mirnawati et al. 2017; Sinurat et al. 2014). The decrease in feed consumption in T5 treatment (30% FPOS) was caused by high crude fiber of 6.8% (Table 2). That is hard for chickens to digest limiting the amount they could eat. Fiber content is known to limit the amount that is consumed (Hidanah et al. 2013). The high crude fiber in the ration makes chickens felt full quickly and therefore its use in poultry feed should be limited at about 3-6% for broiler. The average feed consumption obtained during the study ranged from 2069.1-2292.5g/head/weeks. This result is higher than the feed consumption obtained by Ara et al. (2015) of 2043-2219.3g/head/weeks using *Azolla.*

Administration of FPOS up to the level of 25% of the feed had no significant effect on body weight gain as compared to the control. This result was expected because the feed consumption was not significantly different either. The nutrients and energy consumed is utilized by broiler in the formation of body tissues, so if these are constant, the broiler body weight gain produced will also be the same (Yaophakdee et al. 2018). Thomas et al. (2008). The digestibility of nutrients and the energy utilization from the ration influenced body weight gain, while feed consumption is proportional to increase in body weight (Ara et al. 2015). The smaller body weight gain of broiler fed 30% FPOS (T5) because of the high amount of crude fiber (6.79), which the broiler cannot digest. The amount of this feed that was consumed by the broiler was less. In addition, this high level of the crude fiber reduces the utilization of other nutrients such as protein and results in suboptimal growth (Maslami et al. 2019). Body weight gain is known to decrease with higher crude fiber content (Singh et al. 2017). Mean BWG of broiler obtained during the study ranged from 1289.1-1356.6 g/head/weeks. This result is higher than the BWG of broiler by Supriyati et al. (2015) of 1153-1271g/head/weeks using fermented rice bran (FRB) with *Bacillus amyloliquefaciens* and humic substances.

The results showed that giving FPOS up to 25% in the diet had non-significant effect on the feed conversion. Because the comparison between ration consumption and body weight gain also shows the same result. The quantity of ration was consumed and the amount of weight gain will determine the conversion rate of the ration. According to Ferket and Gernat (2006) the value of feed conversion is influenced by how much of rations consumed and the chicken weight gained. Supriyati et al. (2015) states that the treatment with the same ration consumption and body weight gain will lead to the non-significant conversion of rations.

The significantly lower conversion rate for FPOS of 30% ($P<0.05$) because of the high value of crude fiber resulting in inefficient digestion resulting in reduced weight gain. The feed grade is known to have a significant effect on both consumption and weight gain (Mirnawati et al. 2011). The average FCR obtained during the study was ranged between 1.69-1.89, which was lower than the feed conversion obtained by Ferket and Gernat (2006) of 1.81-1.90 using probiotics supplementations in ration. The results obtained in this study are much lower than those obtained by Mirnawati et al. (2011) which ranges from 1.88-1.91.

Administration of 25% FPOS showed non-significant difference on broiler body weight indicating the nutritional benefits of *P. chrysosporium* and *N. crassa* fermentation in which these microorganisms produce enzymes that change the complex organic compounds in the fiber into materials that were easily digested. Fermentation is known to break down indigestible carbohydrates, fats, proteins, crude fiber or other organic materials increasing digestibility (Brickett et al. 2007). The lower body weight of the broiler fed T5 ration treatment (30% FPOS) because of the high crude fiber in the ration of 6.79%, which exceeds the maximum limit for broiler. The difficulty of digesting a high crude fiber content also results in other food substances being eliminated in the feces without digestion, leading to loss of nutrients reducing body weight (Patel et al. 2015).

In addition, body weight is closely related to feed consumption (517.5-573.1g/head/week) and broiler body weight gain (274.2-336.9g/head/week). The amount of BWG and FBW of broiler was determined by how much feed consumed by broiler. The body weight of broiler obtained during this study was best at 1854.5g/head. This result is lower than the bodyweight of 1797.2g/head obtained by Mirnawati et al. (2018) who used fermented POS with *N. crassa* in diets.

Non-significant difference was found in carcass weights between treatments T1, T2, T3 and T4. Carcass weight is also influenced by body weight, thus large body weight will result in large carcass weight. Zhang et al. (2020) carcass weight was presented as the percentage/relative to live body weight of broiler. Hence, any increase in the absolute carcass weight would not result in significant statistical values as the increased carcass weight was parallel with the increase in final body weight of broiler. The significantly lower conversion rate for FPOS of 30% because it contained the highest amount of fiber-rich FPOS resulted in significantly lower carcass weights, as well as lower body weights as this crude fiber content, exceeds the tolerance limit of crude fiber that can be consumed by broiler. Besides, the carcass weight is affected by palatability. The average weight of broiler carcasses obtained during this study was around 1408.2-1451.2g/head. A higher than mean carcass weight by Akmal and Mairizal, 2013 was found in this study, in which they obtained weights of around 1161.8-1423.7g/head.

There was non-significant difference in carcass percentage between the treatments T1, T2, T3 and T4 as up to 25% FPOS the *Phanerochaete chrysosporium* and *Neurospora crassa* fermentation provided adequate nutritional quality to produce a good amount of muscle. Carcass percentage is also affected by the accumulation of unusable abdominal fat, and the quality of the broiler carcass can be determined by the amount of abdominal fat (Shafey et al. 2014). If the feed contains high metabolic energy but lower amounts of digestible protein, this may result in more abdominal fat to be discarded and a lower quality carcass (Situmorang et al. 2013). The average carcass percentage was obtained during the study was still in the normal range of broiler carcass percentage. The carcass percentage obtained during this study was around 76%. This result is higher than the carcass percentage of 69.38% obtained by Yesuf et al. (2017) who used cassava copra.

The AFP in treatment T1 showed significant differences on treatment T2, T3, T4, and T5. This is due to differences in crude fiber composition in ration. T1 has the lowest fiber content compared to other treatments. This has implications for higher formation of abdominal fat. Okrathok and Khempaka (2020) confirmed giving cassava pulp as a source of dietary fiber can reduce accumulation of abdominal fat and cholesterol in broiler meat. Hidayat et al. (2016) stated that the composition of the ration given to the chicken is one of the many factors affecting the body's fat content. Differences in the percentage of abdominal fat was due to final age of broiler (at 5-6 week of age) so that the fat formed is still tiny. Optimal broiler growth occurs between the ages of 1 day to age 4-6 weeks. In this growth phase, food content absorbed by the livestock body are mostly used for growth, hence no excess of energy (as fat) was stored in the body. According to Salam et al. (2013) in broiler chickens, fat tissue begins to form fast at 6-7 weeks, then at that time, fat accumulation continues to take place more quickly due to abdominal fat so that chicken body weight increases (Diarra et al. 2015; Pratikno 2011). Mean AFP obtained was 1.03% to 1.11%. This result is lower than the abdominal fat percentage of 1.33-1.95 obtained by Fati et al. (2019) who used *Coleus amboinicus* leaf's extract.

Conclusion

From the results of the present study it may be concluded that FPOS with *P. chrysosporium* and *N. crassa* (4:1) may be used 25% in broiler rations, as seen from the FC of 2264g/head, BWG 1289.1g/head, FCR 1.75%, FBW 1854.5g/head, CW 1408.2g/head, CP 76% and AFP 1.04%, which was able to compare the performance of broiler under control ration.

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Author's Contribution

Mirnawati supervised the experiment and writing original manuscript. G. Ciptaan and A. Djulardi conducted the experiment and data analysis. M. Makmur prepared figures and finalize draft. All authors read and confirmed the final version of manuscript.

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