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Performance of quails fed palm oil sludge fermented with *Phanerochaete chrysosporium* and *Neurospora crassa*

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5 Abstract

This study aims to determine what percentage of the use of fermented palm oil sludge (FPOS) with *Phanerochaete chrysosporium* and *Neurospora crassa* in the ration on the performance and quality of quail egg. This study used 200 quails ages 8 weeks, 20 units of 45x20x30 cm battery cages, each unit occupied by 10 quails, feeding and drinking containers. This study uses a completely randomized design experimental method with 5 treatment rations and 4 replications. The treatment rations using 0, 6, 20, 25 and 30% FPOS, respectively. The rations were arranged based on 20% isoprotein and 2800 kcal/kg isocaloric. The variables were observed: feed consumption, egg production (quail day production), egg mass, feed conversion, egg weight, eggshell thickness and yolk egg color. The results of the variance analysis showed that the use of FPOS with *Neurospora crassa* and *Phanerochaete chrysosporium* gave a significant effect ($P > 0.05$) on the feed consumption, egg weight, eggshell thickness and egg yolk color but were significantly effected ($P < 0.05$) on egg production, egg mass and feed conversion. Based on the results of the study it can be concluded that the optimal used of fermented palm oil sludge with *Phanerochaete chrysosporium* and *Neurospora crassa* is 25% in laying quail rations.

Keywords: *Neurospora crassa*, *Phanerochaete chrysosporium*

Introduction

One of the animal that have great potential to be developed is quail. Quail breeding does not require large tracts of land because of its small body, quail has the ability to grow and multiply quickly unlike other birds. From the production, cycle quail are able to lay eggs at the age of 35-42 days and can reach 200-300 eggs per year. In animal husbandry business, feed is a big enough cost, reaching 70-80% of production costs, the high cost of feed is due to some of the ingredients of feed ingredients are still imported such as corn, soybean meal and fish meal. From this problem, we need to find alternative feed ingredients that can reduce feed costs. One that can be used is palm oil sludge (POS), palm oil sludge (POS) is waste generated in the extraction and extortion process of palm fruit taken from Crude Palm Oil (CPO). Palm oil sludge is a potential plantation waste, the price is cheap, does not compete with human needs and does not interfere with animal health.

Based on data released by the Directorate General of Plantations (2016) the area of oil palm plants in Indonesia is around 11,914,499 hectares, with a production of 33,229,381 tons. Each hectare of oil palm can produce 4 tons of oil per year, which is obtained from around 16 tons of fresh fruit bunches (FFB). Every ton of bunches of fresh palm fruit can produce 250 kg of palm oil, 294 kg of palm oil sludge, 35 kg of oil palm flowers and 180 kg of oil palm juice (Mathius et al 2003).

Palm oil sludge has high nutritional content such as: 11.1% crude protein, 17% crude fiber, 12% crude lipid (Noferdiman and Yani 2013), 7.27% hemicellulose and 14.21% lignin (Noferdiman et al 2014). Mirnawati et al (2017) states that palm oil sludge contains 90.47% dry matter, 13% crude protein, 12.31% crude fat and 32.07% crude fiber with a metabolic energy of 1105.87 kcal/kg. Although the nutrient content of palm oil sludge is quite good as a feed ingredient, it is constrained because crude fiber is so high. This is certainly a problem if it is given to poultry because poultry cannot hydrolyze crude fiber. Sinurat et al (2000) stated that palm oil sludge could only be used 5% in poultry rations.

Efforts to reduce the content of crude fiber and increase the use of palm oil sludge in poultry feed need to be processed microbially through fermentation. Fermentation is processing through the process of decomposition or overhaul of food substances from complex forms into simple food substances with the help of enzymes produced by microbes (Mirnawati et al 2019a). Where microbes can degrade lignin and crude fiber so that it is easier to digest and add flavor and aroma and result in an increase the quality and nutritional content of feed ingredients (Mirnawati et al 2019b). Lignin is a lignin degrading enzyme produced by microorganisms that are lignolytic. Where lignocellulose bonds can be broken by ligninases such as lignin peroxidase (LiP), manganese peroxidase (MnP) and laccase (Takano et al 2004). LiP and MnP enzymes are produced by several microorganisms, one of which is *Phanerochaete chrysosporium*.

Howard et al (2003) states that *Phanerochaete chrysosporium* fungi can produce high ligninase and cellulase where *Phanerochaete chrysosporium* can degrade lignin and its derivatives effectively by producing extracellular peroxidase enzymes in the form of lignin peroxidase and manganese peroxidase. Fermentation with *Phanerochaete chrysosporium* in a solid substrate allows a change in components of material that are difficult to digest to be easily digested such as cellulose and hemicellulose into simple sugars thereby increasing the nutritional value of the product. Noferdiman (2009) fermented palm oil sludge (POS) with 6% *Phanerochaete chrysosporium* inoculum for 8 days can reduce crude fiber from 20.16% to 12.22%, lignin from 14.21% to 8.94% and increase in protein roughly from 10.57% to 14.10% but its utilization in broiler rations is only 15%.

Mirnawati et al (2015) conducted a study by selecting three types of *Neurospora crassa*, *Neurospora sitophila* and *Neurospora sp* molds grown on palm oil sludge (POS). The best results in fermented palm oil sludge are *Neurospora crassa* compared to the other two types of mold, where there is an increase in crude protein from 13% to 20.42%, reducing crude fiber from 32.07% to 23.02%, and metabolic energy from 1105 kcal/kg to 2024 kcal/kg. But its utilization in broiler rations is only 22% (Mimawati et al 2018).

To increase the nutrient content and the use of palm oil sludge in poultry rations, the two molds are combined. Mimawati et al (2019c) have fermented palm oil sludge by combining *Phanerochaete chrysosporium* and *Neurospora crassa* with a ratio (4: 1) fermented for 13 days and giving the best results crude protein 27.0%, crude fiber 14.49%, Lignin 14.54%, Caratenoid 2020 g/100g, Nitrogen retention 58.20%, Crude fiber digestibility 57.66%. From the data above there has been an increase in the nutritional content of fermented POS even this FPOS can be used 22% in broiler rations (Mirnawati et al 2018).

Based on the description, it is necessary to do research to find out what percentage (%) of fermented palm oil sludge with *Phanerochaete chrysosporium* and *Neurospora crassa* can be used in quail rations, it is hoped that FPOS can match the quail control ration so as to provide a favorable performance.

Methodology

Purpose of the experiment

This experiment is purposed to study the effect of use several levels of fermented palm oil sludge (FPOS) with *Phanerochaete chrysosporium* and *Neurospora crassa* in the ration on the performance and egg quality of laying quails (feed consumption (g/head/day), egg production (quail day production%), egg mass (g/head/day), feed conversion, egg weight (g/grain), eggshell thickness (mm) and egg yolk colour).

Experimental animal and diet composition

Two hundred laying quails (*Coturnix-coturnix japonica*) aged 8 weeks with an average production of 35%. The experiment was performed in a completely randomized design (CRD) with five treatments in the ration (0%, 10%, 15%, 20%, 25% FPOS) and four replications. There were ten quails per unit of experiment. These laying quails were kept in battery cages (45 x 20 x 30cm). The diets were formulated in iso protein 20% and iso caloric 2800 kcal/kg ration (Djulardi 1995). Feed ingredients used was consisted of yellow maize, soybean meal, rice bran, fermented palm oil sludge (FPOS), fish meal, coconut oil, bone flour, mineral B12 and top mix. Diet and drinking water were provided ad-libitum. The composition of the feed ingredients of treatment ration is shown in Table 1 and the nutrient of the treatment ration is shown in Table 2.

The procedure of preparing FPOS

Fermented palm oil sludge was the product of 80% POS plus 20% rice bran that was fermented with *Phanerochaete chrysosporium* and *Neurospora crassa* as much as 10% with the ratio of each inoculum which is 4: 1. Subsequently incubated in an incubator for 13 days. Furthermore, the FPOS is harvested and roasted at a temperature of 600C to dry. After dry, FPOS is ground and ready to be given in quail ration. Based on the results of Mirnawati et al (2019c), found that the nutritional content of FPOS with combination of *Phanerochaete chrysosporium* and *Neurospora crassa* can be seen in table 3.

Data collection

Data measured are feed consumption, egg production (quail day production), egg mass, feed conversion, egg weight, eggshell thickness and quail egg color

Data analysis

Datas obtained were processed statistically by analysis of variance of a completely randomized design, followed by DMRT test for the difference between treatments, according to Steel and Torrie (1995).

Table 1. Composition of diets

Feed Ingredients	FPOS in diet (%)				
	0	15	20	25	30
Yellow Maize	50.00	45.55	44.00	43.80	43.50
Soybean Meal	23.00	17.75	14.85	12.00	9.00
Rice Bran	7.25	7.00	6.65	5.25	3.75
Fish meal	16.00	16.00	16.00	16.00	16.00
FPOS	0.00	15.00	20.00	25.00	30.00
Coconut oil	1.00	0.95	0.75	0.20	0.00
Bone Flour	1.25	1.25	1.25	1.25	1.25
Mineral B12	1.00	1.00	1.00	1.00	1.00
Top Mix	0.50	0.50	0.50	0.50	0.50
Total (%)	100	100	100	100	100

Table 2. The nutrient content (%) and metabolic energy (kcal/kg) of the treatment diet

Feed nutrition	FPOS in diet (%)				
	0	15	20	25	30
Crude protein	20.03	20.16	20.13	20.12	20.03
Crude Fat	3.13	3.07	2.86	2.29	2.07
Crude Fiber	3.53	4.72	5.30	5.81	6.31
Ca	2.08	2.06	2.04	2.03	2.01
P Available	0.95	0.98	1.00	1.02	1.04
ME	2814	2817	2816	2816	2837

Table 3. The nutrient content (%) and metabolic energy (kcal/kg) of fermented palm oil sludge with *Phanerochaete chrysosporium* and *Neurospora crassa*

Ingredient	Crude Protein	Crude Fiber	Crude Fat	Ca	P	ME
FPOS	26.20	14.49	2.22	0.28	0.65	2788

Result

Mean of feed consumption, egg production (quail day production), egg mass, feed conversion, egg weight, eggshell thickness and egg color of quail that received a mixture of fermented palm oil sludge with *Phanerochaete chrysosporium* and *Neurospora crassa* can be seen in Table 4. The results of the diversity analysis showed that the use of FPOS with *Phanerochaete chrysosporium* and *Neurospora crassa* up to the level of 25% in ration had no significant effect ($P>0.05$) on the feed consumption, egg weight, shell thickness and egg yolk color of laying quail. However, result showed significant effect on egg production, egg mass and feed conversion ($P<0.05$).

Table 4. Mean content of feed consumption, egg production (quail day production), egg mass, feed conversion, egg weight, eggshell thickness and quail egg color

Parameter	FPOS in diet (%)					SEM	p
	0	15	20	25	30		

	0%	15%	20%	25%	30%		
Consumption(g/head/day)							
Feed	22.13	22.13	22.15	22.14	22.84	0.36	0.05
Egg Production (%)	60.39 ^a	60.31 ^a	60.26 ^a	60.21 ^a	58.98 ^b	0.32	0.01
Egg mass(g/head/day)	6.15 ^a	6.13 ^a	6.13 ^a	6.11 ^a	5.98 ^b	0.04	0.01
Feed Conversion	3.60 ^a	3.62 ^a	3.63 ^a	3.66 ^a	3.82 ^b	0.05	0.01
Egg Weight (g)	10.18	10.17	10.16	10.15	10.14	0.01	0.05
Shell Thickness(mm)	0.28	0.28	0.27	0.27	0.26	0.003	0.05
Egg Yolk	6.00	6.25	6.50	6.75	7.00	0.25	0.05

Note: Different superscripts shows significant effect ($P < 0.05$)

Discussion

Results of the research showed that palm oil sludge fermented with *Phanerochaete chrysosporium* and *Neurospora crassa* can be used up to 25% in laying quail rations, as seen from feed consumption, egg production, feed conversion, egg weight, eggshell thickness, and egg yolk color from laying quail that are matching with the control ration.

An increase in the use of fermented palm sludge in quail rations is caused by the palm sludge having undergone fermentation where the fermented product will increase palatability due to physical changes in the fermentation process such as taste, odor, texture and easier to digest from the original ingredients. This is consistent with the opinion of Mirnawati et al (2019b) that the fermented product can produce a better aroma and taste than the original material that are more preferred by the quail. Hidayat (2007) also found that the fermentation process can provide beneficial physical changes such as aroma, taste, texture, which are better than the origin. Murugesan et al (2005) found that fermented products can produce flavors that are preferred by livestock and have several vitamins (B1, B2, B12) so that livestock is more preferred than the original ingredients.

Fermentation can improve the quality of palm oil sludge (POS) and increase digestibility of the ration so that the protein contained in the ration can be utilized for egg production. This is consistent with the opinion of Setiarto et al (2016) that fermentation can increase protein digestibility and amino acid levels, thus with the increase in digestibility of ration and amino acids, egg production will also increase. This is due to the decreasing amount of use of maize, soybean meal and bran and the increasing use of fermented palm oil sludge products so that it still gives the same egg yolk color. FPOS products used in the ration contained 20.25 µg/100g carotenoids (Mirnawati et al 2018). Carotenoids contained in FPOS products come from *Neurospora crassa* which functioned to give the color of egg yolk. According to Hausman and Sandman (2000) β-carotene is a carotenoid group compound that is unstable because it is easily oxidized and will turn into xanthofil. The xanthofil pigment will give the yolk a yellow color. Poultry that consume rations containing higher carotenoid pigments (β-carotene and xanthofil) will produce eggs with a high egg yolk score.

If the use of fermented palm oil sludge in the ration is increased, egg production, egg mass and feed conversion will decrease. The decrease in this parameter is due to increased of crude fiber ration (6.31%). According to Djulardi (1995) that crude fiber in quail feeds is below 5%. For more details, this parameter reduction can be seen in Figures 1, 2 and 3.

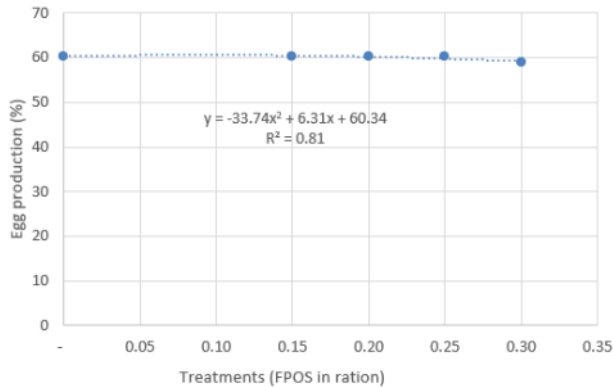


Figure 1. Egg production

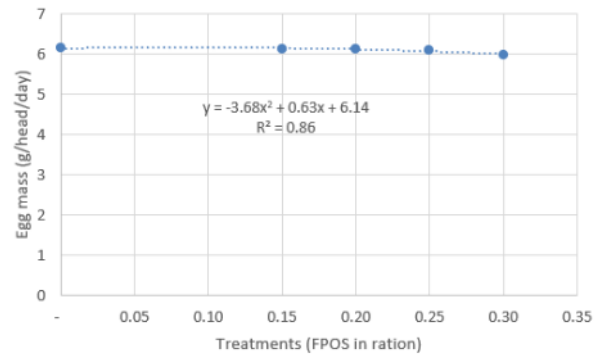


Figure 2. Egg mass

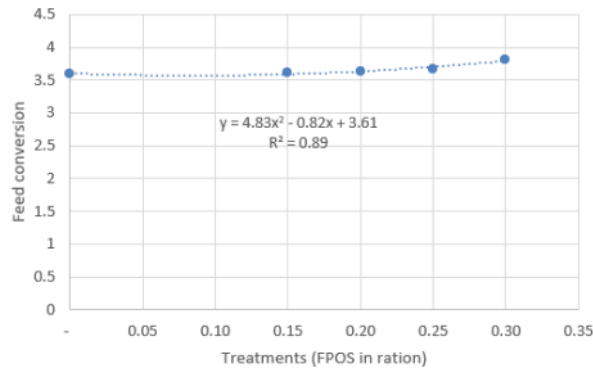


Figure 3. Feed conversion

- From the results of the study, it can be concluded that the optimal use of fermented palm oil sludge with *Phanerochaete chrysosporium* and *Neurospora crassa* is 25% in laying quail rations, it can even reduce the use of soybean meal by about 50% in quail rations.

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