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Research Article

Palm Oil Sludge Fermented with *Lentinus edodes* in the Diet of Broilers

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Abstract

Background and Objective: Palm oil sludge (POS) represents a viable alternative poultry feed component after being fermented by the lignocellulolytic fungi, *Lentinus edodes* (produces ligninase and cellulase), which increases POS nutrient levels and quality. The present study was conducted to evaluate the effect of utilizing POS fermented by *L. edodes* (POSF) in broiler diets on growth performance. **Methodology:** The present study used a completely randomized design (CRD) with five dietary treatment groups (0, 4, 8, 12 and 16% POSF) and four replicates per treatment (5 broilers per replicates). One hundred 3 day-old broiler chicks were included in the study. Diets were isonitrogenous (23% crude protein) and isocaloric (3000 kcal kg⁻¹ diet). The measured parameters were feed consumption, average daily weight gain, feed conversion, carcass percentage and meat cholesterol content. **Results:** Addition of dietary POSF levels up to 16% resulted in good feed consumption, weight gain, carcass percentage and feed conversion but decreased ($p < 0.01$) meat cholesterol content. **Conclusion:** Up to 16% POSF in the diet resulted in good production performance while reducing the cholesterol content of broiler meat, indicating that this feed supplement should be considered for industrial use.

Key words: Palm oil sludge, fermentation, *Lentinus edodes*, broiler performance, meat cholesterol

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Competing Interest: The authors have declared that no competing interest exists.

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

INTRODUCTION

Palm oil sludge (POS) has good potential use as a nonconventional animal feed component, based on its production and nutrient content. Indonesia is currently the largest palm oil producer in the world, generating a total of 22.5 million tons per year¹. Moreover, 70% of the palm oil produced originates from the island of Sumatra. During palm oil production, approximately 45-46% of the material results in waste products, including POS and palm kernel cake¹. The nutrient content of POS includes 11.30% crude protein, 10.43% fat and 1550 kcal kg⁻¹ metabolizable energy^{2,3}.

The problem with utilization of POS is its high crude fiber content (25.80%), which is especially high in cellulose (16.15%) and lignin (19.19%)^{2,3}. Thus, only up to 5% of POS can be included in broiler diets without affecting production and performance^{2,3}. Poultry feed containing a high content of crude fiber (e.g., lignin and cellulose) has low digestibility and its utilization in the diet is limited³. To improve the quality of POS as a poultry feed component, its crude fiber content, particularly lignin and cellulose, must be reduced³. One way to accomplish this is through fermentation with certain species of *Basidiomycetes* white rot fungi that are able to break down all lignocellulose components^{2,3}. In particular, *Lentinus edodes* is a white rot fungi species that has been shown to increase the nutrient content and digestibility value of POS²⁻⁷. *Lentinus edodes* can effectively degrade lignin and cellulose by producing extracellular peroxidase enzymes, such as lignin peroxidase, manganese peroxidase, laccase²⁻⁷ and cellulase^{2,3,5,6}. Furthermore *Lentinus edodes* has been shown to produce eritadenine, which has a hypocholesterolemic effect^{2,3,8-10}.

POS fermented with *Pleurotus ostreatus*, *L. edodes* and *Phanerochaete chrysosporium* can degrade cellulose and lignin while increasing crude protein content, nitrogen retention and metabolizable energy^{2,3}. Our previous research reported that POS fermented with *L. edodes* inoculum dose of 8% and for 9 days increases crude protein content from 11.30-18.36% and decreases crude fiber from 25.80-18.36%².

Hence, the overall dietary quality of this fermented product improved, showing 56.06% nitrogen retention, 48.52% crude fiber digestion, 115.32 µg g⁻¹ eritadenine and 2290.64 kcal kg⁻¹ metabolizable energy². Therefore, the present study evaluated the effect of feeding POSF on broiler production performance and meat quality, especially cholesterol content, as well as its ability to reduce the amounts of other feed components such as corn and soybean meal.

MATERIALS AND METHODS

Fermented feed preparation: *Lentinus edodes* cultures were reconstituted and subcultured using potato dextrose agar for 4 days. Inoculated subcultures were kept at room temperature. POSF was prepared according to the procedure described by Nuraini *et al.*². The substrates contained 80% POS with 20% rice bran and added water (water content 70%). The substrates were autoclaved at 121 °C for 15 min, cooled, inoculated with an 8% inoculum of *L. edodes* and incubated for 9 days. After incubation, the fermented product was dried in an oven at 80 °C for 2 h and 60 °C for 12 h. The dried fermented products were ground in a grinding mill and stored until mixed with the other feedstuffs. Diets were formulated with corn, concentrate BR 131 (Charoen Phokpand production), soybean meal, sheep meat powder, *L. edodes* fermentation products, coconut oil, vitamin mineral mix and CaCO₃.

Experimental design: The present study used a completely randomized design with five dietary treatment groups (0% (control), 4, 8, 12 and 16% POSF) with four replicates per treatment. The treatments reduced corn and soybean meal in the broiler diets. Rations were formulated to be isonitrogenous (23% crude protein) and isocaloric (3000 kcal kg⁻¹ metabolizable energy). The nutrient and metabolizable energy contents of feedstuffs included in the rations are shown in Table 1 and the ration formulations as well as nutrient and metabolizable energy contents are shown in Table 2.

Table 1: Nutrient and metabolizable energy contents of feedstuffs for ration formulation^a

Feedstuffs	Percentage					
	Crude protein	Ether extract	Crude fiber	Ca	P	ME (kcal kg ⁻¹)
Ground yellow corn	8.58	3.77	2.91	0.60	0.10	3370.00
Palm oil sludge fermented ^b	15.56	8.91	14.38	0.21	0.24	2045.99
Soybean meal	45.35	2.49	7.50	0.63	0.36	2240.00
Meat meal	50.00	6.00	2.50	8.00	4.00	2500.00
Concentrate BR 311	22.50	4.40	6.00	0.90	0.60	3200.00
Topmix*	-	-	-	5.38	1.44	-
Palm oil ^b	-	100.00	-	-	-	8600.00

^aLaboratory of feed industry technology, faculty of animal science, university of Andalas, ^bNuraini *et al.*², ^cScott *et al.*¹¹.

Table 2: Ration formulations and their nutrients (%) as well as metabolizable energy contents (kcal kg⁻¹) for broilers*

Feedstuffs	Ration formulations				
	A	B	C	D	E
Ground yellow corn	35.50	31.50	28.25	23.75	19.25
Palm oil sludge fermented	0.00	4.00	8.00	12.00	16.00
Soybean meal	15.00	15.00	15.00	15.00	15.00
Meat meal	11.00	11.00	11.00	11.00	11.00
Concentrate BR 311	40.00	40.00	40.00	40.00	40.00
Topmix	0.50	0.50	0.50	0.50	0.50
Palm oil	0.00	0.50	1.00	2.00	2.80
Total (%)	100.00	100.00	100.00	100.00	100.00
Nutrients and energy					
Crude protein	23.44	23.40	23.08	23.00	23.00
Ether extract	4.08	4.86	5.64	6.89	7.95
Crude fiber	4.68	5.10	5.49	5.90	6.32
Calcium	1.37	1.37	1.37	1.37	1.38
Phosphor	0.77	0.77	0.78	0.78	0.78
ME	3031.90	3011.94	3000.23	3006.57	3000.19

*Calculated based on Table 1

Table 3: Effect of treatment on the feed consumption, average daily weight gain and feed conversion of broilers^{ns}

Treatments	Feed consumption (g/hen/day)	Average daily weight gain (g/hen/day)	Feed conversion
A (0% POSF)	69.05	42.54	1.62
B (4% POSF)	68.73	42.05	1.63
C (8% POSF)	68.32	42.67	1.60
D (12% POSF)	69.23	41.80	1.66
E (16% POSF)	68.91	42.23	1.63
SE	0.20	0.49	0.02

ns: Not significantly different (p>0.05), SE: Standard error of the mean, POSF: Palm oil sludge fermented with *Lentinus edodes*

Birds and housing: One hundred 3 day-old CP 707 broiler chicks were included in the present study (5 broilers in each box; 20 broilers in each treatment group). Chicks were individually weighed and randomly allotted to each of the five POSF treatment group. All chicks were provided with uniform feeder and water space with house boxes reared under standard management conditions throughout the 4 week experimental period. Feed and water were provided *ad libitum*.

Variables: The parameters measured were feed consumption (g/head/day), average daily weight gain (g/head/day), feed conversion, carcass percentage and meat cholesterol content (mg/100 g).

Data analysis: All of the data were statistically analyzed by one-way analysis of variance in the completely randomized design (CRD). Significant differences between treatments were determined by using Duncan's multiple range test with a p<0.05 considered significantly different.

RESULTS

The effects of each dietary treatment on the performance of broilers are presented in Table 3.

Feed consumption: The feed consumption of broilers was not significantly affected (p>0.05) by inclusion of POSF in the diet. Feed consumption in the 0% POSF treatment was 69.35 g/hen/day and increasing utilization of POSF until 16% resulted in similar feed consumption (68.23 g/hen/day).

Average daily weight gain: Inclusion of POSF in the diet of broilers did not significantly influence (p>0.05) their average daily weight gain. The average daily weight gain in the control treatment (0% POSF) was 42.54 g/hen/day and increasing utilization of POSF until 16% resulted in a similar effect (no negative effect) on the average daily weight gain (42.67 g/hen/day).

Feed conversion: The feed conversion ratio of broilers was not significantly affected (p>0.05) by inclusion of POSF in the diet. The feed conversion with the 0% POSF treatment (1.62) was not different from that with the 16% POSF treatment (1.63).

Carcass percentage: The carcass percentage of broilers was not significantly affected (p>0.05) by supplementation with POSF in the diet. The carcass percentage in the control treatment (71.28%) was similar to that in the 16% POSF treatment (71.23%).

Table 4: Effect of treatment on the carcass percentage and meat cholesterol^{ns}

Treatments	Carcass (%)	Meat cholesterol (mg/100 g)
A (0% POSF)	1.78	100.02 ^a
B (4% POSF)	72.00	94.05 ^b
C (8% POSF)	71.60	90.24 ^c
D (12% POSF)	70.28	85.15 ^d
E (16% POSF)	71.23	78.70 ^e
SE	1.61	1.39

ns: Not significantly different ($p > 0.05$), SE: Standard error of the mean, POSF: Palm oil sludge fermented with *Lentinus edodes*

Table 5: Comparison of amino acid content in palm oil sludge (POS) to POS fermented with *Lentinus edodes* (POSF)

Amino acids	Amino acids concentration (%)	
	POS	POSF
Aspartate	1.18	1.21
Glutamate	2.52	2.50
Serine	0.54	0.55
Histidine	0.49	0.50
Glycine	0.38	0.35
Threonine	0.51	0.57
Arginine	0.62	0.60
Alanine	0.98	1.12
Tyrosine	0.19	0.25
Methionine	0.50	0.54
Valine	0.43	0.48
Phenylalanine	0.52	0.49
Iso leucine	0.39	0.43
Leucine	1.12	1.06
Lysine	1.06	1.12
Proline	0.27	0.29
Cysteine	0.51	0.56

POSF: Palm oil sludge fermented with *Lentinus edodes*

Meat cholesterol: Inclusion of POSF in the diet of broilers significantly decreased ($p < 0.05$) the meat cholesterol content in a concentration-dependent manner. Increasing the amount of POSF decreased the meat cholesterol content. The meat cholesterol in the 0% POSF treatment (78.70 mg kg⁻¹) was decreased compared to that in the 16% POSF treatment (100.02 mg kg⁻¹) (Table 4).

DISCUSSION

Using up to 16% POSF in the diet of broiler chickens had no significant effect on any of the measured production performance parameters, except the cholesterol content in the meat. In fact, increasing utilization of POSF until 16% resulted in similar feed consumption. Utilization up to 16% POSF (with the reduction of corn and soybean meal content) was just as palatable as the 0% POSF (more utilization of corn and soybean meal content) in the diet. Fermented products are preferred by poultry compared to the non-fermented material¹⁰, due to good flavor. Moreover, fermented products

also increase amino acids and introduce additional vitamins (e.g., vitamins B1, B2 and B12). Table 5 shows that amino acids in POSF increased compared to non-fermented product. The feed consumption of broilers given rations with POSF in the present study ranged from 68.3-69.23 g/hen/day. Results of present study were lower than that reported by Aljubori *et al.*¹², who showed that broiler feed consumption is 72.96 g/hen/day when fed 10% fermented canola and lower than that reported by Nuraini *et al.*² (72.95 g/bird/day) when broilers fed 20% palm kernel cake fermented with *Lentinus edodes*.

The similar average daily weight gain and carcass percentage of the 16 and 0% (control) POSF groups was most likely caused by the similar feed consumption. Similar feed consumption indicates that the amount of nutrients consumed and digested are also similar, especially protein and energy, resulting in similar daily body weights and carcass percentages. Body weight is affected by the nutrient content in the diet and feed intake^{13,14}. The average daily weight gain observed in the present study (42.23 g/hen/day) was lower than that observed by Aljubori *et al.*¹² (46.25 g/bird/day) in broilers fed 10% fermented canola and lower than that reported by Nuraini *et al.*² (45.68 g/bird/day) in broilers fed 20% palm kernel cake fermented with *Lentinus edodes* for 4 weeks.

Furthermore, feed conversion ratios for all POSF treatment groups were similar because there was no significant change in feed consumption or average daily weight gain. The feed conversion ratio is the ratio between feed intake and the amount of meat produced^{8,13}. Feed conversion can be used as an indication of the production coefficient where a lower value indicates more efficient use of feed to produce meat. In the present study, feed conversion was defined as the ratio between feed intake and weight gain. The similar feed conversion ratio between 0% POSF (1.62) and 16% POSF treatment group observed in the present study, demonstrated that substitution of up to 16% POSF for corn and soybean meal in the diet of broilers does not significantly affect feed intake, average daily weight gain or meat production. The average feed conversion ratio in the present study was higher than that reported by Nuraini *et al.*² (1.60) in broilers fed 20% fermented palm kernel cake and higher than that presented by Aljubori *et al.*¹² (1.58) in broilers fed 10% fermented canola.

Interestingly, the meat cholesterol content decreased to 21.32% (from 100.02 mg/100 g in the 0% POSF treatment to 78.70 mg/100 g in the 16% POSF treatment), which may have been attributed to the increased amount of the fermentation product, eritadenine, in the broiler diet. Eritadenine is a

product of *L. edodes* fermentation with known hypocholesterolemic effects^{7,8,9}. Adenosine derivatives of eritadenine [2(R), 3(R)-dihydroxy-4-(9-adenyl)-butyric acid, lentinacin and lentysine) have been isolated and identified as active hypocholesterolemic components of shiitake mushroom. Eritadenine reduces serum cholesterol levels in mice by accelerating the excretion of ingested cholesterol and its metabolic decomposition¹². Furthermore, eritadenine not only affects the metabolism of cholesterol but also the metabolism of phospholipids and fatty acids in rats⁷.

CONCLUSION

Inclusion up to 16% POSF in broiler diets did not significantly affect feed consumption (68.91 g/hen/day), average daily weight gain (42.23 g/hen/day), feed conversion ratio (1.63), or carcass percentage (71.23%), but it reduced the meat cholesterol content by 21.32%.

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SIGNIFICANCE STATEMENT

The present study evaluated the effect of using POSF as an alternative feed component on broiler production performance. This result will aid the development of POSF formulations for broiler rations, which will not only provide good performance (non-negative effect) and increase meat quality but also reduce costs for farmers.

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