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Research Article

The Effects of Adding Phytase Enzymes and Lemuru (*Sardinella* sp.) Oil to Rice Bran on the Meat Quality of ISA Brown Chickens (*Gallus gallus domesticus*)

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Abstract

Background and Objective: Enzyme phytase and lemuru (*Sardinella* sp.) oil supplementation may increase the meat quality of ISA Brown Chickens (*Gallus gallus domesticus*). This study aimed to determine the effects of adding phytase enzymes to feed on the quality of chicken meat by analyzing protein, fat, cholesterol, high-density lipoprotein (HDL) and low-density lipoprotein (LDL) content. **Materials and Methods:** Twenty-four month-old ISA Brown Chickens were allocated six different feed formulations with four replications as follows: (T1) commercial feed (control), (T2) commercial feed +2% lemuruoil, (T3) 95% commercial feed +5% rice bran +1% phytase enzymes +2% lemuruoil, (T4) 90% commercial feed +10% rice bran+1% phytase enzymes+2% lemuruoil, (T5) 85% commercial feed +15% rice bran +1% phytase enzymes+2% lemuruoil, (T5) 85% commercial feed +15% rice bran +1% phytase enzymes+2% lemuruoil, (T5) 85% commercial feed +15% rice bran +1% phytase enzymes+2% lemuruoil, (T5) 85% commercial feed +15% rice bran +1% phytase enzymes+2% lemuruoil. The experimental measurements included protein, fat, cholesterol, LDL and HDL. Data were statistically analyzed using one-way ANOVA. **Results:** Chickens fed 80% commercial feed with 20% rice bran with the addition of 1% phytase enzymes and 2% lemuru oil had the lowest cholesterol (33.81 mg dL⁻¹) and LDL (18.23 mg dL⁻¹) levels compared to those fed the others formulations. This group also had the highest HDL content (139.90 mg dL⁻¹); fat and protein content were not significantly different. **Conclusion:** Commercial feed consisting of 20% rice bran with the addition of 1% phytase enzymes meat quality of ISA Brown Chickens.

Key words: ISA Brown Chicken, lemuru, meat quality, phytase enzymes, rice bran

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

INTRODUCTION

In Indonesia, plants, such as rice bran, are often used as feed for ISA Brown Chickens (*Gallus gallus domesticus*)^{1,2}. Rice bran is cheap, easy to obtain and rich in nutrients, such as protein, zinc, iron and folic acid³. Unfortunately, rice bran contains phytic acid, an anti-nutrient that prevent poultry from absorbing nutrients properly⁴.

Phytic acid can reduce protein solubility because it easily reacts with proteins to form phytate-protein complexes⁵. The formation of phytate-mineral compounds or insoluble proteins can reduce the availability of minerals and the nutritional value of protein rations⁶. Phytic acid is a secondary compound in plants in the form of the main deposits of phosphorus in grains, accounting for approximately 60-80% of the total phosphorus and phytic acid molecules containing high P minerals, which is approximately 28.8%⁷.

Phytic acid can strongly bind twovalent cations, such as Ca^{2+} , Mg^{2+} , Zn^{2+} and Fe^{2+} and can bind starches, proteins and amino acids so that they cannot be digested^{8,9}. Supplementation with phytase enzymes can suppress the negative effects of phytate in feed. Phytase is an enzyme that belongs to the phosphatase group, which is able to hydrolyze phytic compounds in the form of Myo-inositol (1, 2, 3, 4, 5, 6) hexa phosphatase to myo-inositol and organic phosphate¹⁰.

This study aimed to determine the effect of adding phytase enzymes in feed on the quality of chicken meat, in addition to 2% lemuru oil (*Sardinella* sp.). Lemuru fish oil derived from marine fish is a source of polyunsaturated fatty acids (PUFAs), which contain many long-chain fatty acids such as omega-3 and omega-6¹¹. Omega-3s can improve meat quality by reducing LDL and increasing HDL^{12,13}. The quality of chicken meat was analyzed by measuring the values of protein, fat, cholesterol, HDL and LDL.

MATERIALS AND METHODS

Animals and diets: These experiments were conducted at the animal houses of the Animal Nutrition Laboratory of Faculty of Veterinary Medicine Universitas Airlangga and Testing Service Unit Faculty of Pharmacy Universitas Airlangga. The phytase enzymes were obtained from commercial products and the lemuru (*Sardinella* sp.) oil was sourced from the Sardinella Processing Factory in Banyuwangi, East Java, Indonesia.

Twenty-four month-old ISA Brown Chickens were included in this study. Six treatments using four chickens each were evaluated using a completely randomized design. The treatments in this study were as follows: (1) Commercial feed (control), (2) commercial feed + 2% lemuru oil, (3) 95% commercial feed + 5% rice bran + 1% phytase enzymes + 2% lemuru oil, (4) 90% commercial feed + 10% rice bran + 1% phytase enzymes + 2% lemuru oil, (5) 85% commercial feed + 15% rice bran + 1% phytase enzymes +2% lemuru oil and (6) 80% commercial feed + 20% rice bran + 1% phytase enzymes + 2% lemuru oil. Feeding and drinking were carried out twice a day *ad libitum* (7AM and 4PM). Feeding was carried out during the study for 35 days.

The average dry matter, ash, crude protein, ether extract, crude fiber, organic matter, nitrogen-free extract, calcium and phosphorus in treatment feed with the combination of phytase enzymes are shown in Table 1.

Data collection and meat quality analysis: After 35 days, protein, fat, cholesterol, HDL and LDL were analyzed.

Experimental design: A completely randomized design was used. Meat quality (protein, fat, cholesterol, HDL and LDL) was statistically assessed by one-way ANOVA (SPSS Version 22).

Table 1: Effect of varving levels of phytase enzy	ymes and 2% lemuru oil supplementation to feed on	chemical composition

Nutrition (%)	Treatments							
	1	2	3	4	5	6		
Dry matter	90.19	91.49	91.19	90.56	89.85	90.08		
Ash	16.18	15.94	14.90	16.66	16.92	17.31		
Crude protein	18.19	18.60	18.74	18.77	18.70	18.78		
Ether extract	5.40	5.52	6.71	6.61	6.75	7.06		
Crude fiber	5.17	6.09	6.28	6.54	7.57	8.08		
Organic matter	74.01	75.55	76.29	73.90	72.94	72.77		
Nitrogen-free Extract	45.26	45.54	44.36	42.39	40.25	38.95		
Calcium	5.78	6.09	5.83	5.87	6.33	6.37		
Phosphorus	0.50	0.50	0.55	0.60	0.65	0.90		
Metabolite energy (kcal kg ⁻¹)	2671.79	2700.24	2760.76	2662.55	2596.72	2582.81		

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Parameters	Treatments	Treatments							
	1	2	3	4	5	6			
Protein meat (%)	18.27ª	19.39 ^b	19.51 ^b	20.47 ^c	19.95 ^{b,c}	19.69 ^{b,c}			
Fat meat (%)	3.23ª	2.80 ^b	2.91 ^{bc}	2.57°	2.52°	2.48 ^c			
Cholesterol (mg dL ⁻¹)	190.01ª	138.32 ^b	134.07 ^b	84.30 ^c	41.04 ^c	33.80 ^c			
HDL (mg dL ^{-1})	58.75°	72.30 ^{b,c}	90.12 ^{a-c}	112.04 ^{a,b}	120.34 ^{a,b}	139.90ª			
LDL (mg dL ^{-1})	67.08 ^a	33.75 ^b	29.57 ^b	37.99 ^b	36.01 ^b	18.23 ^b			

Table 2: Effect of supplementation of enzyme mixture and 2% lemuru oil on protein, fat and cholesterol of chicken meat

Different superscripts showed significant differences (p<0.05)

The difference between means was analyzed using Duncan's multiple-range test to compare the means within the fixed factors¹⁴.

RESULTS AND DISCUSSION

The results of the obtained data on meat protein, fat, cholesterol, HDL and LDL of chicken meat were presented in Table 2.

Across the 35 days of this study, cholesterol, HDL and LDL significantly differed; however, there were no significant differences in protein and fat content. Treatment 6 produced the most favorable results with the lowest levels of cholesterol and LDL.

Supplementation of phytase enzymes and lemuruoil in commercial feed did not significantly affect the protein and fat content of meat quality. This result was likely due to the presence of phytate content in the rice bran that was not well degraded by the phytase enzymes.

Rice bran was used as feed due to its low price compared to commercial feed. However, rice bran contains phytate, an anti-nutrient similar to tann in and protease inhibitors that produce a complex form of essential amino acids, enzymes, proteins and binding minerals that disrupt nutritional value and protein digestibility¹⁵. Unfortunately, the phytase enzyme used in this study did not degrade the phytate well due it slow concentration, which was only 1%. Phytase enzymes hydrolyze the phytate myo-inositol hexaphosphate (IP6) substrate to its lower ester to increase absorption before entering the duodenum^{16.}

A significant effect was observed on cholesterol levels due to the addition of lemuru fish oil to the feed. Lemuru oil contains mono-unsaturated fatty acids (MUFAs) and PUFAs, which can reduce LDL content in the blood and increase HDL¹¹. According to a previous study, cholesterol was the most significant compound often found in large steroid families such as lemuru, in nerve, brain, and blood tissues but not in plant tissues and vegetable products¹⁷.

The cholesterol in living organisms come from two sources, either produced by themselves (endogenous

cholesterol) or from foods (exogenous cholesterol); in this case, lemuruoil produced the cholesterol¹⁸. Lemuru oil functions as a source of unsaturated fatty acids and reduces cholesterol, which consisted of saturated fatty acids, by improving blood glucose control^{19,20}. This study agrees with the findings of Engel and Tholstrup²¹ who reported that adding phytase enzymes and lemuru oil in the diet significantly decreased cholesterol, LDL and increased HDL in the blood²¹.

SIGNIFICANCE STATEMENT

This study identified possible synergistic effects of adding 1% phytase enzymes and 2% lemuru oil in a 20% rice bran combination that can be beneficial for enhancing the meat quality of ISA Brown Chickens. This study will help researcher sun cover critical areas of rice bran utilization in animal feed and potentially open new areas of research, thus stimulating a new theory on this enzyme-oil combination and possibly other combinations as well.

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