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## **Performance and blood lipid profile of ducks (*Anas sp.*) fed a ration containing calcium soap made from palm fatty acid distillate (PFAD)**

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### **Abstract**

The research was conducted to examine the effect of fed with calcium soap made from palm fatty acid distillate (Ca-PFAD) in the ration on the performance and blood lipid profile of male bali ducks (*Anas sp.*). The design used in this study was a completely randomized design with 4 treatments and 6 replications, and each experimental unit used 10 male bali ducks aged two weeks with homogeneous body weight. The four types of treatment, namely: A: ration without added Ca-PFAD as a control; B: feed with the addition of 1.5% Ca-PFAD; C: feed with the addition of 3.0% Ca-PFAD; and D: feed with the addition of 4.5% Ca-PFAD, respectively. The rations were prepared isocalorically (ME: 2900 kcal/kg) and isoprotein (CP:17%). The results showed that the administration of 1.5-4.5% Ca-PFAD in the diet significantly ( $P < 0.05$ ) increased feed consumption, final body weight, body weight gain, dry matter and organic matter digestibility, and feed efficiency compared to control. On the other hand, decreased significantly ( $P < 0.05$ ) on total cholesterol, total pad-fat, and abdominal fat in ducks. It can be concluded that 1.5-4.5% Ca-PFAD in feed can improve performance of male bali and decreased total cholesterol concentration in serum of bali ducks.

**Keywords:** Palm fatty acid distillate, growth, cholesterol, ducks

### **Introduction**

The by-product of processing palm oil into cooking oil will be obtained in the form of palm oil fatty acid distillate (PFAD). Calcium soap made from PFAD (Ca-PFAD) is rich in vitamin E, especially tocotrienols, phytosterols, and squalene.

The abundant availability of PFAD has the potential as an animal feed ingredient, in addition to its low price, it also does not compete with ingredients for food, such as palm oil. Palm oil is one of the most widely consumed vegetable oils in the world, next to soybean oil, canola oil and sunflower seed oil. One of the largest producers of palm oil in the world is Indonesia, while the largest consumer is India [1].

Palm oil contains minor components, such as carotenoids, vitamin E (tocopherols and tocotrienols), phytosterols, squalene, and phenolic compounds [2, 3, 4]. The refining process will produce refined palm oil as the main product and palm fatty acid distillate (PFAD) as a by-product that has not been widely used [6]. Palm fatty acid distillate (PFAD) contains very high free fatty acids and is often used in the manufacture of soaps and detergents. The addition of calcium is intended so that the fat given as a supplement to livestock does not interfere with the digestion of livestock [6]. PFAD can be used as raw material for animal feed supplements after being reacted with calcium in the form of protected fat. Serves as an energy source for livestock and has a positive effect on milk production, as well as cow fertility. Giving 0.45 kg/day calcium soap to cows can increase milk production by 3-8% [5].

Fatty acids need to be converted into soap before consumption, because direct consumption of fatty acids by livestock can interfere with the activity of digestive bacteria in the rumen of livestock. PFAD is a valuable source of bioactive compounds wherein the unsaponifiable fraction (USF) of PFAD contains vitamin E, phytosterols, and squalene, and USF is a potential candidate for dietary supplements and all bioactive compounds in USF are expected to act synergistically [7]. According to [8], saponification of PFAD produces unsaponifiable matter (USM) which is rich in vitamin E, especially tocotrienols, phytosterols, and squalene. Total cholesterol and LDL cholesterol of rats fed USM-fortified diet were lower than controls, as well as better ratios of total cholesterol to HDL and LDL cholesterol to HDL.

Mice that were given additional phytosterols in a high-fat diet were found to have lower levels of total cholesterol, LDL cholesterol, and triglycerides than controls [9].

Based on the description above, the research was conducted to examine the effect of calcium soap (Ca-PFAD) in the ration on the performance and carcass of male Bali ducks.

## Materials and Methods

### Animal treatments and experimental design

The design used in this study was a completely randomized design with 4 treatments and 6 replications, and each experimental unit used 10 male Bali ducks aged two weeks with homogeneous body weight. The rations given to the treatment ducks were complete rations in the form of mash prepared using ingredients, such as yellow corn, pollard, soybeans, fish meal, NaCl, and minerals. The rations were given according to the need for ducks aged 2-8 weeks according to the [10]. The rations were prepared isocalorically (ME: 2900 kcal/kg) with isoprotein (CP: 17%). The drinking water provided during this study was taken from the local drinking water company (PAM). The four types of treatment, namely: A: ration without added Ca-PFAD as a control; B: feed with the addition of 1.5% Ca-PFAD; C: feed with the addition of 3.0% Ca-PFAD; and D: feed with the addition of 4.5% Ca-PFAD, respectively. Each cages of 200×100×50 cm<sup>3</sup> (length × width × height) with 10 birds to each cage.

### Live performance

Treatment rations were given twice a day. Feed consumption (FI) was calculated every week. Weighing of body weight (BW) of ducks was carried out every week and before weighing the ducks were fasted to eat for 12 hours, while drinking water was still given. Weight gain (LWG) is the difference between final body weight and initial body weight. Feed conversion ratio is the ratio between the amount of feed consumed and the weight gain of ducks in the same unit (FI: LWG).

### Retention and nutrient digestibility

To measure feed digestibility, the total collection method was carried out for three days. Ducks were placed in individual cages with limited feeding of 100 g/head/day, for three days. Feeding is done twice a day. Under the individual cages, a plastic tray is placed to accommodate the feces for three days. Samples of feed and feces were then dried in an oven at 75 °C until a constant weight (DW) was obtained. Then the sample was ground and in the oven again at a temperature of 105 °C until a constant weight (DM) was obtained. The sample was

then finely ground with a diameter of 1 mm and analyzed for dry matter content (DM), organic matter (OM), according to procedure [11]. All tests were carried out in triplicate.

**Calcium Soap (Ca-PFAD):** Calcium soap used in this study was in the form of mash. The chemical composition of the calcium soap used in the study were: 7.85% water; 54.24% BETN; 97.32% TDN; 18.6% ash; 0.8% crude protein; 31.3% crude fat; 3.0% Calcium; and a gross energy of 6562 kcal/kg<sup>[5]</sup>. The physical form of calcium soap is presented in Figure 1.



Fig 1: Physical form of calcium soap (Ca-PFAD)

**Blood lipid profile:** Cholesterol analysis used the Lieberman-Burchard method, using 5 cc of duck blood taken at the base of the duck's wing at the end of the study. The sterol solution in chloroform is reacted with concentrated sulfuric anhydrous acetic acid. This test produces a color from bluish green to green, depending on the cholesterol content of the sample. The resulting solution is stamped on a spectrophotometer to obtain the optical density (DO). The results were then compared with the DO of the standard solution.

### Statistical analysis

All research data were analyzed with one-way variance. If there was a significant difference ( $P < 0.05$ ) between treatments, it was continued with Duncan's multiple distance test.

### Result

Table 1 presents data on ration consumption, protein and energy consumption, BW, LWG, and feed efficiency in ducks for 42 days of observation. The average feed consumption, protein and energy in the treatment of ducks B, C, and D increased ( $P < 0.05$ ) rather than control. Ducks treatments B, C, and D consumed each ration: 15.30%; 15.89%; and 15.55% significantly ( $P < 0.05$ ) higher than the control (A).

Table 1: Performance of male Bali duck (*Anas sp.*) aged 8 weeks fed calcium soap powder (Ca-PFAD)

Variables	Treatments <sup>1</sup>				SEM <sup>2</sup>
	A	B	C	D	
Feed intake (g/bird/days)	72,61b	83,72a	84,15a	83,90a	2,491
Feed consumption (g/bird/42 days)	3049,70b	3516,18a	3534,43a	3523,97a	120,26
Protein consumption (g/bird/42 days)	489,17b	562,94a	566,57a	564,19a	15,714
Energy consumption (kcal/bird/42 days)	8847,18b	10196,92a	10260,45a	10233,61a	351,85
Initial body weight (g)	180,37a	181,04a	180,81a	180,25a	3,961
Final live weight (g)	1074,71b <sup>3</sup>	1283,29a	1275,06a	1254,63a	42,063
LWG (g/42 days))	894,34b	1102,25a	1094,25a	1074,38a	40,287
Feed conversion ratio	3,41a	3,19b	3,23b	3,28b	0,032
Dry matter digestibility (%)	67,05a	70,81b	69,95b	69,83b	0,618
Organic matter digestibility (%)	68,59a	72,04b	71,87b	71,62b	0,803

1. Ca-PFAD levels in the diet, namely: 0% (A); 1.5% (B); 3.0% (C); and 4.5% (D), respectively.

2. Different notations <sup>a,b</sup> on the same line, showed significantly different ( $P < 0.05$ ).

The use of 1.5-4.5% Calcium soap in the diet significantly ( $P < 0.05$ ) increased the final body weight and weight gain of ducks. The body weight of treatment group B, C, and D increased, respectively: 19.41%; 18.64%; and 16.74% higher ( $P < 0.05$ ) compared to control (A). Likewise, the weight gain of ducks during 42 days of observation in treatment B, C, and D ducks, respectively: 23.25%; 22.35%; and 20.13% significantly ( $P < 0.05$ ) higher than the treatment duck group A.

Dry matter digestibility (DM) and organic matter (OM) of feed increased ( $P < 0.05$ ) by supplementing of Calcium soap (Ca-PFAD) in the ration (Table 1). Dry matter digestibility in treatment duck groups B, C, and D, respectively were: 5.61%; 4.33%; and 4.15% significantly ( $P < 0.05$ ) higher than the control. Likewise, the digestibility of organic matter in groups B, C, and D ducks increased significantly ( $P < 0.05$ ), respectively: 5.03%; 4.78%; and 4.42% higher than the control.

Administration of 1.5-4.5% Calcium soap (Ca-PFAD) in duck rations for 6 weeks of observation resulted in a significant reduction ( $P < 0.05$ ) in the content of pad-fat and abdominal fat (Table 2). The average number of pad fat in the treatment group B, C, and D ducks, respectively: 19.15%; 27.65%; and 23.40% significantly ( $P < 0.05$ ) lower than the control. Likewise, the content of abdominal fat in groups B, C, and D ducks, respectively: 16.67%; 12.50%; and 15.28% significantly ( $P < 0.05$ ) lower than the control.

The use of 1.5-4.5% Calcium soap (Ca-PFAD) in duck rations for 6 weeks of the experiment, did not effect ( $P > 0.05$ ) on the content of triglycerides, high-density lipoprotein (HDL), and low-density lipoprotein (LDL) in duck blood serum (Table 2). However, significantly ( $P < 0.05$ ) decreased the total cholesterol concentration in duck blood serum. The concentration of total cholesterol in the blood serum of treatment B, C, and D ducks, respectively: 9.20%; 9.00%; and 11.45% lower ( $P < 0.05$ ) than the control.

**Table 2:** Blood lipid profile of male Bali ducks fed a diet containing Calcium soap made from PFAD (Ca-PFAD).

Variable	Treatments <sup>1</sup>				SEM <sup>2</sup>
	A	B	C	D	
Pad-fat (% berat badan)	0.47a	0.38b	0.34b	0.36b	0.029
Abdominal-fat (% berat badan)	0.72a	0.64b	0.61b	0.63b	0.032
<b>Blood lipid profile</b>					Normal
Total cholesterol (mg/dL)	175.92a	159.73b	160.08b	155.47b	<200
Triglycerides (mg/dL)	58.16a	51.62a	48.94a	50.03a	<150
HDL (mg/dL)	85.93a	92.76a	89.24a	87.28a	>40
LDL (mg/dL)	51.85a	49.36a	45.51a	42.74a	<130

1. Ca-PFAD levels in the diet, namely: 0% (A); 1.5% (B); 3.0% (C); and 4.5% (D), respectively.

2. Different notations <sup>a,b</sup> on the same line, showed significantly different ( $P < 0.05$ ).

## Discussion

The use of Calcium soap made from PFAD (Ca-PFAD) at the level of 1.5-4.5% in the ration can increase final body weight and weight gain. Fatty acids need to be converted into soap before consumption, because direct consumption of fatty acids by livestock can interfere with the activity of digestive bacteria in the rumen of ruminants and the digestive tract of poultry<sup>[7]</sup>. PFAD can be used as raw material for animal feed supplements after being reacted with calcium in the form of protected fat. Serves as a source of energy for livestock and has a positive effect on livestock productivity.

The increased weight gain of ducks in the presence of Calcium soap in the ration was due to the presence of phytochemical compounds in PFAD. According to<sup>[12]</sup>, palm oil contains 50% saturated fatty acids, 40% monounsaturated fatty acids, and 10% polyunsaturated fatty acids, as well as sufficient beta-carotene, tocopherol and tocotrienols, all of which is a natural antioxidant. It was also reported that the components of phytosterols, squalene, coenzymes, polyphenols, phospholipids, quinones, ubiquinones, coumarins, and amino acids were needed by ducks to support their growth. The addition of fat to the diet, in addition to supplying energy, also increases the absorption of fat-soluble vitamins, provides varying amounts of essential fatty acids, reduces dust levels, increases feed palatability, and increases energy efficiency<sup>[13, 14, 15, 16]</sup>. Similar to the research of Monfaredi *et al.* (2011) that there is an increase in body weight and feed efficiency with the addition of vegetable oil or animal oil into broiler chicken rations. Conflicting results were reported by<sup>[17]</sup>, that the use of 1-3% palm oil in laying hens rations significantly reduces feed consumption. Also reported by<sup>[18]</sup> that the addition of Calcium-palm fatty acid

(Ca-PFAD) in feed up to 15% significantly reduced feed consumption and had no effect on weight gain, carcass, and physical composition of broiler carcasses. However, it markedly increased unsaturated fatty acids, omega-3 fatty acids, and decreased body fat in broilers.

The increase in feed efficiency in ducks with the presence of Calcium soap in the ration was due to the fact that Ca-PFAD can act as a source of energy, vitamins, and other phytochemical compounds. According to<sup>[19]</sup>, fat plays an important role in poultry nutrition, because fat contains at least twice the available carbohydrate and protein energy, so fat is widely used in broiler rations to meet energy needs. Chickens fed a diet containing oil showed better performance and higher fat digestibility than rations fed without oil intake, even though the nutritional value of the food was the same<sup>[20]</sup>. According to<sup>[13]</sup>, fat can reduce the rate of digesta flow in the duck's digestive tract, thus providing space for longer nutrient absorption so that feed efficiency increases.

Monfaredi *et al.*<sup>[15]</sup> stated that there was an increase in body weight and feed efficiency with the addition of vegetable oil or animal oil into broiler chicken rations. Calcium soap made from PFAD is an energy source feed ingredient with a metabolized energy content of 6562 kcal/kg<sup>[5]</sup>. Giachetto *et al.*<sup>[21]</sup> evaluated the performance and hormonal profile of broilers fed rations with various energy levels, and found that appropriate energy levels in the ration increased ration consumption and feed efficiency.

Poorghasemi *et al.*<sup>[20]</sup> stated that chicken rations fed with oil-containing feed showed better performance and higher fat digestibility than rations fed without oil, even though the nutritional value of the food was the same. Vegetable fat, high in essential fatty acids. As reported by<sup>[22]</sup>, that linoleic and

linolenic fatty acids derived from lin-seed oil are recognized to be very important metabolically for broilers, because broilers are not able to synthesize these fatty acids, and linoleic acid is the only fatty acid. essential fats that need to be present in the ration. According to [17], palm oil is able to affect the transit of feed in the digestive tract of laying hens, so that it can increase digestibility and absorption of nutrients, so that the production performance of laying hens increases in rations containing up to 2% palm oil. Liang *et al.* [15] reported that the use of soybean oil in the diet can significantly improve the performance and local immune system in the small intestine of broilers.

Mineral Ca is always associated with phosphorus in the Ca: P ratio [24]. Collett [25] suggested that high concentrations of Ca in poultry diets can impair the quality of manure by increasing the moisture content of manure. In addition, poor litter quality is often associated with the incidence of foot pad dermatitis (FPD), which has received increasing attention in the broiler industry [26]. The incidence of foot pad dermatitis (FPD) will decrease when broiler chickens are fed a diet containing sufficient Ca [26]. Calcium has important biological functions and must be provided in adequate amounts. Inadequate Ca intake can affect bone mineral content, muscle function, and other body mineral functions [27]. Chickens fed a diet containing 1.25% Ca showed higher body weight gain and nutrient digestibility compared to 1.5%. In addition, different types of ration oils have different effects on growth performance, intestinal morphology, and digestibility of mineral nutrients in broiler chickens [19].

The accumulation of large amounts of fat in the abdominal cavity (abdominal-fat) is a problem in modern broiler strains. Abdominal fat is removed by evisceration of the abdominal cavity, thereby reducing the yield of processing or post-harvest poultry [15]. The use of 1-3% Ca-PFAD in the diet significantly reduced pad fat and abdominal fat. This is due to the tocotrienol fraction in PFAD which can function as a natural antioxidant [32], and suppress the formation of triglycerides. The type of fat or the same fat profile used in the preparation of the ration also greatly influences the fat accumulation in the duck's body. As reported by [28], that the quality of poultry carcass meat is influenced by the fatty acid profile of the food consumed by broilers during rearing. Soybean oil supplementation in diets with low energy content can improve performance and reduce belly fat, serum cholesterol, and LDL levels compared to chickens receiving rations supplemented with beef fat or tallow [15].

Kolani *et al.* [17] stated that supplementation of 1-3% palm oil in the diet had no effect on the amount of abdominal fat, but significantly reduced liver weight. It was also reported that the use of 3% palm oil in the diet significantly reduced blood serum cholesterol levels of laying hens, while serum triglyceride and glucose levels did not show any significant difference. The results showed that supplementation of broiler rations with 40 g soybean oil/kg feed significantly improved performance and reduced serum cholesterol, LDL, and belly fat compared to chicks receiving rations containing beef fat [15].

Total cholesterol levels and the amount of abdominal fat in ducks that received rations with 1-3% calcium soap supplementation decreased significantly. This is due to the content of phytosterols and squalene in PFAD. Squalene as one of the bioactive compounds in PFAD has anti-cancer and cholesterol-lowering properties [3]. Squalene has the ability to

increase the fecal excretion of bile acids which causes a decrease in blood cholesterol levels. Squalene has also been reported to reduce cholesterol levels in plasma [29], inhibit the formation of atherosclerosis, and reduce cholesterol levels in the liver [30]. Besides squalene, PFAD is also high in vitamin E, and its phytosterols have a potential hypocholesterolemic effect. According to [31], vitamin E as an antioxidant prevents the oxidation of LDL cholesterol in an *in vitro* study. The tocotrienol-rich fraction of PFAD has antioxidant and cholesterol-lowering properties, and has hepatoprotector, immunomodulatory, antioxidant properties, and lactogenic effects in animal test experiments [7]. Tocotrienol compounds in PFAD are able to inhibit HMG Co-A reductase, the enzyme responsible for converting HMG to mevalonate in cholesterol biosynthesis [8]. Phytosterols have the ability to compete with cholesterol during the formation of micelles in the intestine. Palm oil is known to contain sufficient amounts of beta-carotenoids [32, 33, 34, 35].

### Conclusions

It can be concluded that 1.5-4.5% Calcium soap made from PFAD (Ca-PFAD) in feed can increase dry matter and organic matter digestibility, weight gain and feed efficiency in male bali ducks aged 8 weeks. Pad-fat deposition, abdominal fat, and total cholesterol levels in blood serum of bali ducks decreased significantly in the presence of Ca-PFAD in the diet at the level of 1.5-4.5%.

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