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Performance and Egg Quality of Laying Chickens Feed Supplementation with *Plectranthus amboinicus* Lour Spreng Leaf Flour

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Abstract. This study aims to examine the potential of *Plectranthus amboinicus* Lour Spreng leaf flour PLP as a supplement to the performance of laying hens. 30 laying hens aged 22 weeks were used and divided into 5 treatments and six replications. The five treatments were T0 (fed without PLP), T1, T2, T3, and T4 namely the group that was fed with PLP 2.5% in a row, 5%, 7.5% and 10%. The study was conducted for six weeks. Parameters measured were weekly body weight, feed efficiency, the weight and present of carcass and meat cholesterol. Another parameter were egg quality which includes egg weight and cholesterol levels. One of the biochemical parameters measured is the level of uric acid in the blood. At the end of the study, the chickens were slaughtered to obtain blood for the measurement of uric acid, as well as the measurement of carcass quality. The data obtained were analyzed using one-way ANOVA at a significance level of 95%. The results showed that PLP supplementation in the feed reduced body weight and increased feed efficiency significantly ($p < 0.05$) and also reduce meat cholesterol. The addition of PLP in the feed did not affect egg weight but decreased yolk cholesterol levels and uric acid significantly if compare to tha control group.

INTRODUCTION

In recent years, there has been an increased consumer demand for meat and egg products that focuses on animal welfare during production and product safety and quality. As such, feeding forage or pasture intake in outdoor-based systems (conventional free-range systems or organic systems) for laying hens has gained tremendous attention in the poultry research field, as highlighted by certain reviews. For this purpose some vegetables can be used for . *Plectranthus amboinicus* Lour is a vegetable that is rich in fiber which is expected to be used as a chicken feed supplement to improve chicken performance.

Plectranthus amboinicus Lour Spreng leaves are used to treat cholesterol disease [1]. This plant contain flavonoids such as apigenin, quercetin and salfigenin which have many properties. Plants containing flavonoids are efficacious in lowering cholesterol [2]. Where flavonoids are able to erode cholesterol deposits on the walls of coronary blood vessels, so they will not trigger other diseases caused by cholesterol, such as hypertension, stroke and heart disease. Quercetin is a flavonoid compound and has strong antioxidant activity [3]. Flavonoids and tannins are antioxidants that can fight LDL cholesterol and prevent damage to blood vessel tissue [4]. Flavonoids work as inhibitors of the HMG-CoA reductase enzyme so that cholesterol synthesis decreases. Meanwhile, tannins inhibit fat absorption in the intestine by reacting with protein). mucosa and intestinal epithelial cells. *Plectranthus amboinicus* leaves are also used as cough medicine, sore throat, fever, nasal congestion, hepatopathy, kidney stones, asthma,

bronchitis, helmianthiasis, colic convulsions and epilepsy [5]. Biological activity of *Plectranthus amboinicus* L. Spreng plant, namely as antimicrobial, anti-inflammatory, hypocholesterolemic, hepatoprotective, anti-androgenic, antioxidant, anti-tumor, anti-dabetic, analgesic, diuretic, anti-carcinogenic and vasodilator [6]. According to previous researchers [7] that the higher the application of PLP, the lower the liver weight and levels meat cholesterol. Supplementation of 20% PLP flour in the feed did not affect broiler body weight. . The purpose of this study was to examine the effect of feed supplementation with leaf flour (*Plectranthus amboiniciu* L. Spreng (PLP) on the performance and egg quality of laying hens. In addition, the level of uric acid in the blood was also observed.

MATERIALS AND METHODS

Plectranthus amboinicus Leave Powder

The leaves of *Plectranthus amboinicus* were obtained from self-planting located in the yard of the Medan State University Animal House. For the manufacture of flour used the method used by [8]. In detail it can be explained as follows. The leaves are washed and picked and then drained for one night. Furthermore, the leaves are finely sliced and dried in the sun or in the oven at 40°C. The process of drying the leaves for 14 days, until the leaves look dry and brittle. Next, the dried leaves are blended until smooth and then sieved using a sieve or calico cloth. . *Plectranthus amboinicus* leaves that have become flour are stored at room temperature and closed containers. The total need for flour for 35 days is 4,515 g.

Commercial Feed

The feed used in this study was commercial feed type 324-2 specifically for chickens that produce eggs. The feed was obtained from one of the livestock shops located at the MMTC market in Medan. The need for commercial feed type 324-2 specifically for chickens that produce eggs during the acclimatization period of 36 weeks to the last trial period at 42 weeks of age can be calculated based on the need for chicken feed reduced by feed shrinkage. During the productive period, chickens need 85 g of feed/head/day while there is a decrease of 2%, so to get the right feed, $2/100 \times 85 = 1.7$ g. From the results obtained, the total feed needed is 86.7 g / head / day during the experimental period ends, therefore the total commercial feed for 35 days is 86.520 g.

Supplementation of *Plectranthus amboinicus* Leaf Flour in Commercial Feed

PLP supplementation with commercial feed. PLP was weighed according to the required dose. An example for 10% PLP supplementation treatment can be seen in the following example:

$$10\% = 10/100 \times 86,7 = 8,6 \text{ g PLP /day/head} + 78,1 \text{ g commercial feed/day/head.}$$

If there are 6 chickens in the PLP supplementation treatment with a concentration of 10% then:

$$\begin{aligned} 10\% &= 8,6 \text{ g PLP} + 78,1 \text{ g commercial feed times } 6 \\ &= 51,6 \text{ g TBB} + 468,6 \text{ g commercial feed} \end{aligned}$$

So that 51,6 g of PLP + 468,6 g commercial feed mixed and stirred until homogeneous.

Layer Chicken Coop

5 cages are needed with 1 cage containing 6 plots. Each plot of the cage is 40 cm long, 40 cm wide, and 40 cm high. The height of the cage from the ground is 60 cm. Each plot consists of 1 chicken. The cage is made of wood. Prior to use, the cage was sterilized using alcohol for 3 days. Each plot is equipped with a place to eat and a place to drink. The red color of the feed is more attractive than other colors so that the chicken becomes appetizing [9]. Then, 3 plots were given one 40 watt incandescent lamp as heating and lighting that stimulated the growth of chickens.

Laying Hens

The chickens used in this study were arabian laying hens as many as 30 tails obtained from the Suwandy Deli Serdang Chicken Farm. The Arabic chicken used was 40 weeks old and had laid two eggs. The first week of arriving at the cage, acclimatization was carried out.

Experimental Design

After acclimatization for one week, the chickens were first weighed and their body weight recorded, the chickens were weighed only at the beginning of the weekly study, and at the end of the study. Furthermore, they were grouped into five treatment groups and each group consisted of six chickens. The five treatments are:

- T₀ = feed without PLP supplementation
- T₁ = feed with 2.5% PLP supplementation
- T₂ = feed with 5% PLP supplementation
- T₃ = feed with 7.5% PLP supplementation
- T₄ = feed with 10% PLP supplementation

The treatment of feeding supplementation with PLP was for 35 days. Laying hens are fed every day by the everyday basis method.

Data Collection Technique

The data collected in this study consisted of Body Weight, Feed Efficiency, Quality of Carcass, Egg Weight, Yolk Cholesterol and Uric Acid. Body weight measurements were carried out every week during the study, which was six weeks, using an analytical balance with an accuracy of 0.1 g. Each chicken was weighed by placing it on a scale and then recording its weight. Feed efficiency is determined based on consumption and feed conversion. Feed consumption is obtained by using the following formula:

$$\text{Feed consumption (g/head/day)} = \frac{\text{weigh of feed given-remaining food}}{7 \text{ days}} \quad (1)$$

While the feed conversion is obtained by using the formula:

$$\text{Food Conversion} = \frac{\text{total feed consumption}}{\text{total body weight}} \quad (2)$$

Carcass quality measured was carcass weight and percent using formula:

$$\text{Carcass weight} = \text{Cutting weight} - \text{by weight of feathers, head, legs and offal} \quad (3)$$

$$\text{Carcass Quality} = \frac{\text{weight of carcass}}{\text{cutting weight}} \times 100 \% \quad (4)[10]$$

Measurement of egg quality in this study included egg weight and egg yolk cholesterol which was carried out every week. The method of measuring egg weight is by weighing the eggs of each chicken in each treatment, then averaged. Measurement of egg cholesterol using a spectrophotometer at a wavelength of 630 nm. The cholesterol content (mg/100 g) in the sample was obtained by the formula (5) [11]:

$$\text{Cholesterol} = \frac{\text{sample absorbance}}{\text{standard absorbance}} \times 200 \text{ (standard concentration)} \quad (5)$$

Data analysis

The data obtained were tabulated and analyzed using one way Anova. The mean difference of each treatment was tested with Duncan's New Multiple Range test (DMRT).

RESULT AND DISCUSSION

Chicken Performance

The performance of chickens measured in this study were weekly chicken weight, carcass quality, meat cholesterol content and feed efficiency (feed consumption and feed conversion).

Body Weight Weekly

Weekly body weight can be seen in Figure 1. In the figure it can be seen that from the first week of treatment to week five, all groups of chickens fed PLP supplementation lost weight significantly compared to the control group. However, from week five to week six, the weight of the T₁, T₂ and T₄ treated chickens increased and the T₃ group experienced a significant decrease.

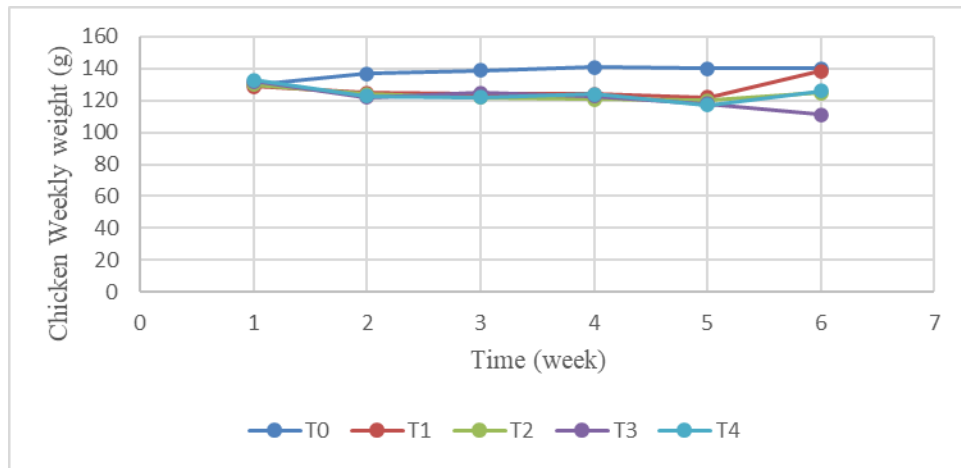


FIGURE 1. The effect of feed supplemented PLP on laying hens Weekly Average Weight (n = 30) T₀ is the group of chickens without PLP supplementation. T₁ T₂ T₃ and T₄ are groups of chickens fed PLP 2.5, 5, 7.5 and 10 percent supplementation respectively

The weight loss in this study is the same as the study conducted by [12]. In this study, it was explained that supplementation of 3% and 4% PLP in chicken feed reduced the weight of broiler chickens significantly compared to the control group. Supplementation of feed with PLP causes palatability of broiler chickens to feed along with increasing levels of PLP. The palatability of the feed decreased due to the sensation of bitter taste and odor in the feed. Another factor that causes a decrease in chicken weight is the presence of linalool in *Plectranthus amboinicus* [13] Linalool plays a role in increasing the activity of hepatic microsomal enzymes thereby increasing the metabolic rate.

Another factor that affects this weekly weight loss is because PLP contains saponins. This is similar to the results of the study [14] which added 1% and 2% bay leaf flour to the feed. The results showed that the live weight of chickens fed with 1% and 2% bay leaf flour did not increase body weight significantly compared to the control. And it was explained that the possible cause was due to the presence of saponins and tannins in bay leaves. Saponins are thought to cause a decrease in growth by reducing ration consumption. Saponins in tea leaves can inhibit growth because it reduces ration consumption. This decrease in ration consumption was thought to be caused by a delay in the rate of cache emptying due to the presence of saponins [14].

Quality of Carcass

Other chicken performances measured in this study were weight and carcass percent (Figure 2). Carcass weight is the cut weight without head, legs, off and fur. In Figure 2 it can be seen that the carcass weight decreased significantly in the group of chickens fed PLP supplementation. The highest decrease was in the feed treatment group with 2.5% PLP supplementation. Although the carcass weight decreased, the carcass percentage increased in the groups of chickens fed 5% and 7.5% PLP supplementation, respectively.

The results of this study are in line with the previously research [7] which stated that 20% PLP supplementation in feed reduced carcass weight significantly compared to controls. The percentage of rust in this study increased significantly in the PLP 5% and 7.5% treatment. The increase in carcass percentage in this study was higher than the study [7]. The percentage of carcasses in this study was lower than the results of the study¹⁵, which explained that the percentage of carcasses with water extract of *Phyllanthus niruri* L and Moringa (*Moringa oleifera* Lam) was 69.43% - 72.98%. the carcass percentage value in this study was still classified as normal, where the carcass percentage in broiler chickens was from 65 - 75% of body weight.

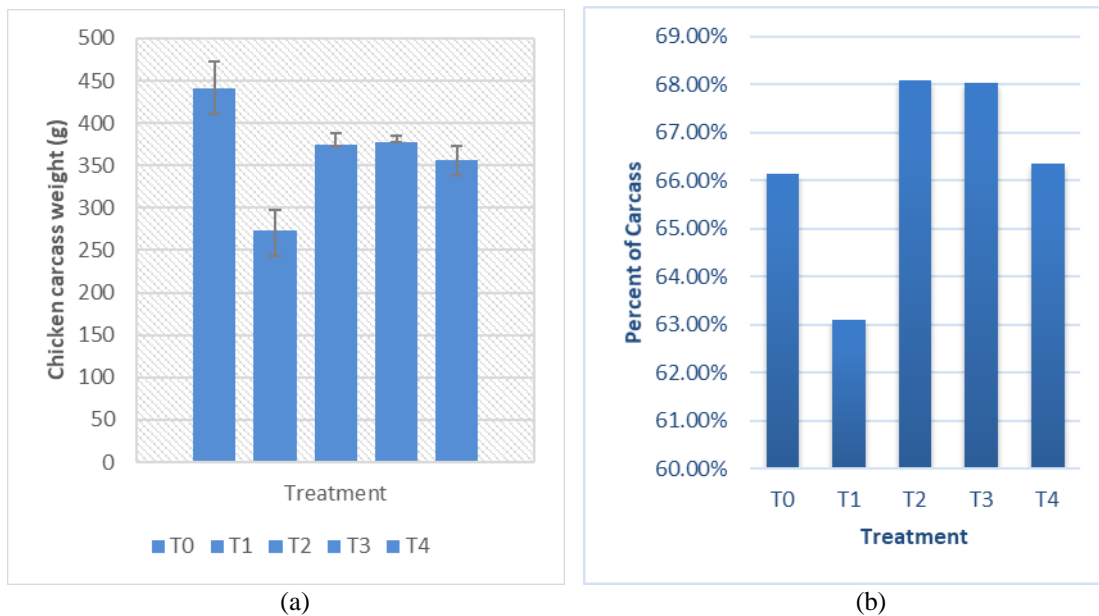


FIGURE 2. The effect of feed supplemented PLP on laying hens Chickend carcass weight (a) and Percent of carcass (b) (mean + STDEV; n = 30); T₀ is the group of chickens without PLP supplementation. T₁, T₂, T₃, and T₄ are groups of chickens fed PLP 2.5, 5, 7.5 and 10 percent supplementation respectively

Meat Cholesterol

Feeding PLP supplementation in this study significantly reduced cholesterol in meat compared to the control group. In Figure 3 it can be seen that the higher the PLP supplied in the feed, the higher the decrease in meat cholesterol. The decrease in cholesterol in meat is due to the presence of saponins contained in PLP. The powder leaf of *Plectranthus amboinicus* (Lour.) Spreng also contained non-volatile compounds which had been reported by previous studies such as flavonoids, terpenoids, saponins, steroids, tannins, proteins, and carbohydrates [16-18]. Saponins are natural detergents found in many plants. Saponins have detergent or surfactant properties because they contain both water-soluble and fat-soluble components. They consist of a fat-soluble nucleus, having either a steroid or triterpenoid structure, with one or more side chains of water-soluble carbohydrates. Many authors have studied the biological properties of saponins on animal and human nutrition [19,20]. It has been reported that saponins when supplemented in diets reduce cholesterol content in blood and tissues in monogastric mammals, such as rats, gerbils and humans. In addition to saponins, according to [21], one of the secondary metabolites contained in PLP is flavonoids. Plants containing flavonoid compounds are efficacious in lowering cholesterol.

Utilization of hypocholesterolemic sources such as saponins from dried alfafa has been carried out by [22] who showed that with the addition of alfafa saponins it can reduce breast meat cholesterol and total chicken fat. Diosgenin (steroid saponin) is a very useful compound to control hypercholesterolemia by inhibiting cholesterol absorption and increasing cholesterol secretion [23]. Saponins remain within the gastrointestinal tract. Some interact directly with cholesterol producing an insoluble complex which prevents cholesterol absorption [24]. Others appear to affect cholesterol metabolism indirectly by interacting with bile acids and increased faecal excretion of bile acids is observed in response to feeding saponins of this type [25].

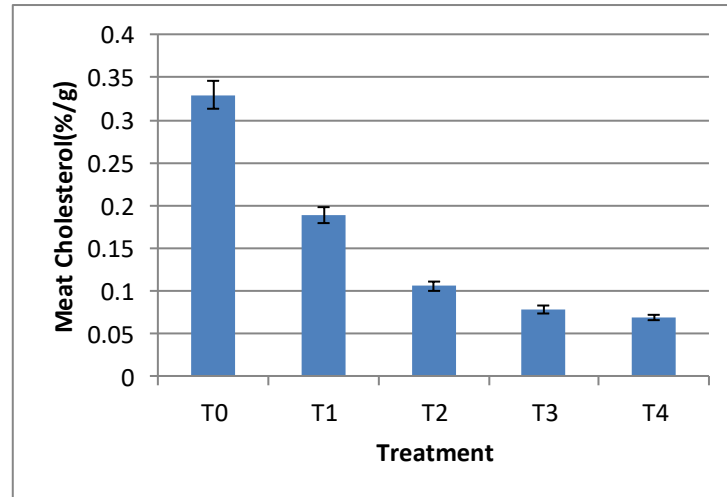


FIGURE 3. The effect of feed supplemented PLP on laying hens Meat Cholesterol (mean + STDEV; n = 30); T₀ is the group of chickens without PLP supplementation. T₁, T₂, T₃, and T₄ are groups of chickens fed PLP 2.5, 5, 7.5 and 10 percent supplementation respectively

Supplemented 0.08% and 0.12% alfalfa saponins to broilers diets could significantly decrease cholesterol content of serum and liver ($P < 0.05$), increased HDL-C content in serum and bile acids content in feces ($P < 0.05$), highly decreased LDL-C content and HMG-CoA reductase mRNA abundance ($P < 0.01$), implicating alfalfa saponins could block enterohepatic circulation of bile acids, thus suppress the synthesis of cholesterol to some extent [26]. 0.08% alfalfa saponins also could significantly reduce insulin content in serum ($P < 0.05$). Added moderate alfalfa saponins to broilers feed could reduce TG content and Hydrocortisone content in serum to a certain extent, effectively improve cholesterol metabolism.

In addition, the addition of fibrous feed in chicken rations also aims to make the fat and cholesterol in the chicken's body can be removed through excreta by binding most of the bile salts. The release of some bile salts stimulates the body to synthesize bile salts from the body's cholesterol so that the cholesterol in the body as a whole can be reduced.

Feed Consumption

Feed consumption is an indicator to determine the feed consumed in sufficient quantities and in accordance with the needs of Arab chickens. The amount of feed given cannot be all consumed, therefore the calculation of feed consumption needs to be done. Feed consumption can be calculated every day where the ratio between the amount of feed given to the amount of feed remaining. The amount of feed consumed by Arabian chickens aged 0.8-1.5 years is 85 grams/day [27]. In this study, PLP supplementation had no effect on chicken feed consumption for six consecutive weeks with $F_{hit} = 0.50; 2.57; 0.80; 0.01; 0.42$ and 0.30 and p value = $0.73; 0.06; 0.53; 0.99; 0.78$ and 0.87 . The feed consumption did not differ in this study because the use of PLP at the level of 0% - 10% was still tolerable for chicken consumption (Figure 4).

A low feed conversion value can increase the efficiency of the use of feed and conversely a high feed conversion value can reduce the efficiency of feed use [28]. The productive period of Arabian chicken is 0.8-1.5 years and has a body weight of 2 kg and requires 85 grams of feed/day, So that the conversion of the ration obtained is around 0.0429 [29].

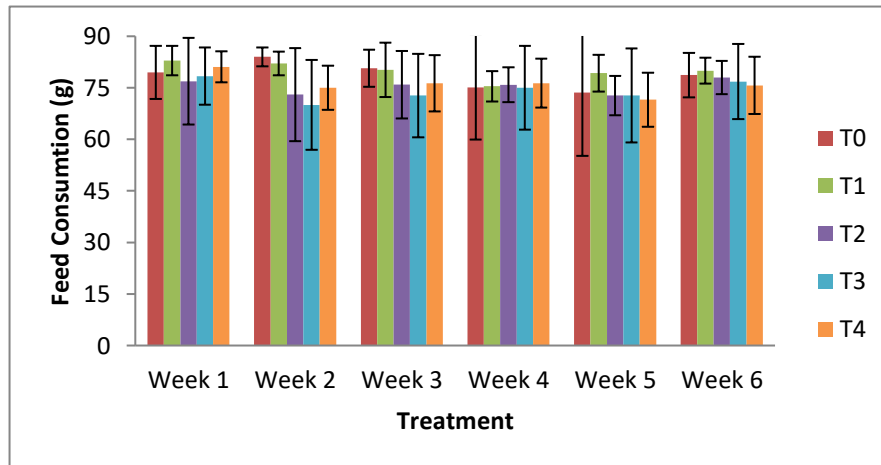


FIGURE 4. The effect of feed supplemented PLP on laying hens Feed Consumption (mean + STDEV; n = 30); T₀ is the group of chickens without PLP supplementation. T₁, T₂, T₃, and T₄ are groups of chickens fed PLP 2.5, 5, 7.5 and 10 percent supplementation respectively

Feed Conversion

Conversion of ration is the ratio between the amount of ration consumed with body weight gain in a certain period of time. The greater the feed conversion rate, the less economical the use of the feed is, on the contrary, the smaller the conversion rate, the more economical it is. Feed conversion of laying hens ranges from 2.1-2.3 [30].

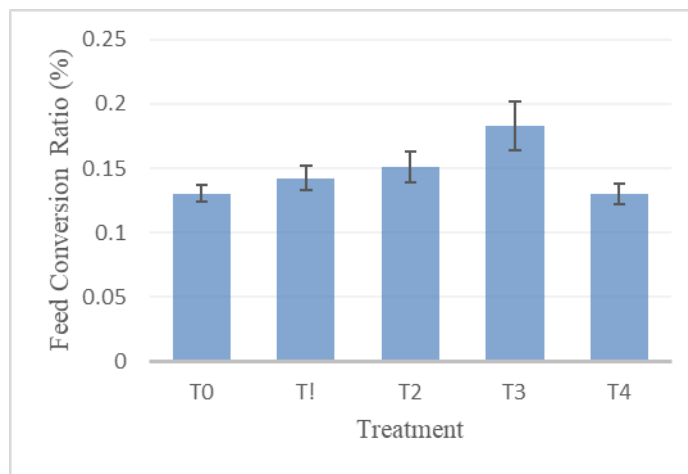


FIGURE 5. The effect of feed supplemented PLP on laying hens Feed Conversion (mean + STDEV; n = 30); T₀ is the group of chickens without PLP supplementation. T₁, T₂, T₃, and T₄ are groups of chickens fed PLP 2.5, 5, 7.5 and 10 percent supplementation respectively

In this study, the greater the concentration of PFL in the feed, the higher the feed conversion. Based on data analysis using one-way Annava shows that the use of PLP in feed has a significant effect on increasing feed conversion with Fhit value = 2.81; p= 0.047 (Figure 5). The results of the LSD test showed that the administration of PLP was 0%; 5%; 7.5% significantly different from PLP 10%. Several factors that affect feed conversion in chickens are health status, food quality and rearing management. The highest feed conversion in this study was the group of chickens with 7.5% PLP supplementation. Increased feed conversion in this study.

Egg Quality

The quality of eggs studied in this study was egg yolk cholesterol levels and egg weight. The results of the study are explained as follows

Yolk Cholesterol

Egg yolk cholesterol levels can be seen in Figure 4 generally high especially in egg yolks. In this study, at the beginning of the study or the first week of the study, it can be seen that chicken egg cholesterol in general is still high, with an average of over 300 mg/100 g (Figure 6). However, cholesterol levels decreased after being given PLP supplementation feed. The decline started at week two of the study and continued to decline until week six. In the picture it can also be seen that the higher the percentage of PLP levels supplemented in the feed, the higher the decrease in cholesterol. The cholesterol content in 1 large egg is 213 mg [31].

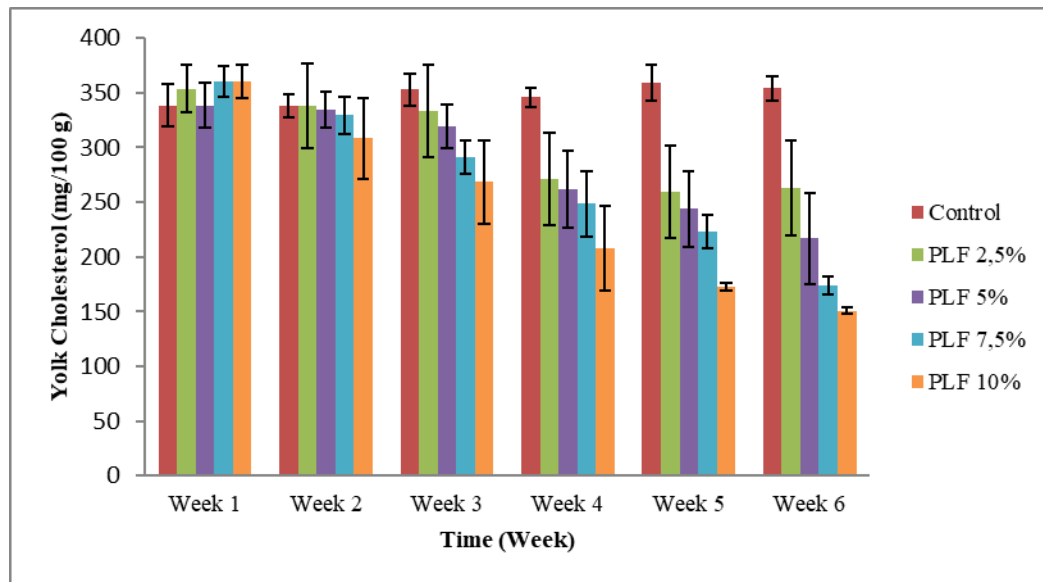


FIGURE 6. The effect of feed supplemented PLP on laying hens Yolk Cholesterol (mean + STDEV; n = 30); T₀ is the group of chickens without PLP supplementation. T₁, T₂, T₃ and T₄ are groups of chickens fed PLP 2.5, 5, 7.5 and 10 percent supplementation respectively

Egg Yolk Cholesterol

Sources of cholesterol in the body are from food intake, and denovo synthesis in the body. The regulation of cholesterol levels can be caused by the intake of fiber in the feed. The decrease in cholesterol levels in egg yolks was due to the antihyperlipidemic properties of *Plectranthus amboinicus* L. This was explained by [21] that administration of ethanolic extract of *Plectranthus amboinicus* leaves 200 mg/kg body weight of alloxan-induced diabetic rats, increased the lipase enzyme in serum so that decreased total cholesterol and triglyceride levels and increased HDL, total protein and calcium significantly compared to diabetic rats. In addition, PLP has a high fiber content which can bind cholesterol in the digestive tract so that the external source of cholesterol decreases.

The Phytochemical database [32] reported that *Plectranthus amboinicus* leaves contain beta carotene and niacin. High consumption of beta carotene can reduce cholesterol levels in egg yolks, because beta carotene inhibits the action of the enzyme HMG-CoA reductase (Hydroksi methyl glutaryl-CoA) which plays a role in the formation of mevalonic. Mevalonic is needed in the biosynthesis of cholesterol in the liver and egg yolk cholesterol is synthesized in the liver so that inhibition of the HMG-Coa enzyme by beta carotene can reduce cholesterol synthesis in the liver by [33]. Quercetin contained in *Plectranthus amboinicus* Utilization of quercetin by laying hens improves their antioxidant status, reduces yolk [34] and serum cholesterol levels [35].

Egg Weight

In this study, egg weights were weighed every week for six weeks. The results of data analysis showed that there was no significant difference ($p>0.05$) between the egg weights of the control group and those given PLP. The average weight of Arabic chicken eggs is 40 grams/eggs [36]. It can be seen from Figure 7 that the egg weight in the study is still within the standard average weight of chicken eggs. The results of this study are in line with research [37] which said that the provision of papaya leaf flour in the ration did not have a significant effect on egg weight. Furthermore, it was explained that good ration palatability did not affect egg weight. Factors that determine the weight of an egg are the environment, age of the parent, and egg composition [38].

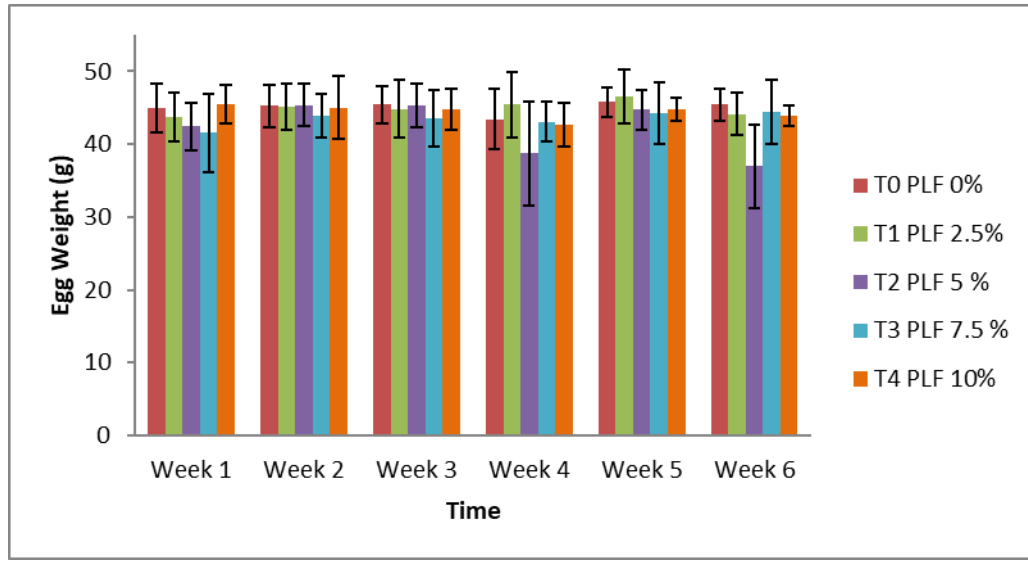


FIGURE 7. The effect of feed supplemented PLP on laying hens Egg Weight (mean + STDEV; $n = 30$); T₀ is the group of chickens without PLP supplementation. T₁, T₂, T₃, and T₄ are groups of chickens fed PLP 2.5, 5, 7.5 and 10 percent supplementation respectively

Uric Acid

Poultry is indeed a uricotelic animal, which is an animal that produces nitrogenous waste in the form of uric acid. Therefore, for gout sufferers, they should avoid consuming chicken meat. In this study, the group of chickens fed PFL supplementation had a significant decrease in uric acid levels compared to the control group (Figure 8). In this study, uric acid levels in control chickens were high, above 10 mg/L. However, in all groups of chickens that were given PLP supplementation, their uric acid levels decreased significantly compared to the control group.

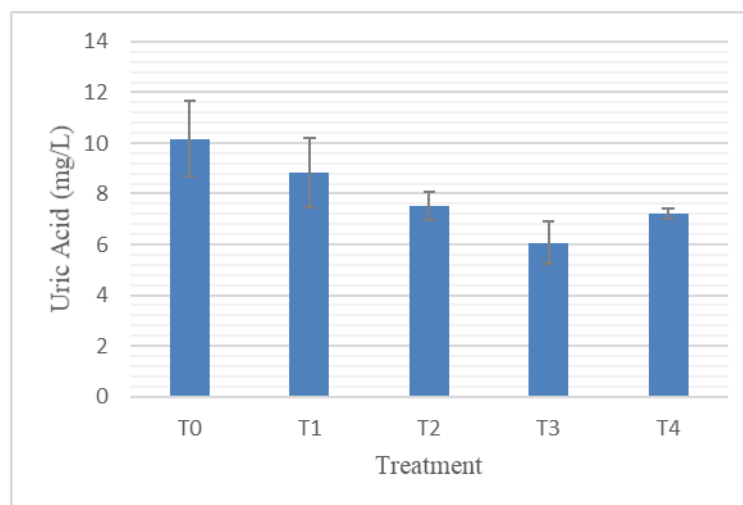


FIGURE 8. The effect of feed supplemented PLP on laying hens uric acid (mean + STDEV; n = 30); T₀ is the group of chickens without PLP supplementation. T₁ T₂ T₃ and T₄ are groups of chickens fed PLP 2.5, 5, 7.5 and 10 percent supplementation respectively

Plectranthus amboinicus leaf extract contains phenols, flavonoids, alkaloids, and saponins [39] which have been proven to be anti-convulsant in experimental animals and contain compounds of carene, terpinene, camphor, and carvacrol which have anti-rheumatoid function [40].

More specifically, the bioactive compounds of plant-based functional foods are divided into six categories, namely flavonoids, phenolic acids, alkaloids, saponins, polysaccharides, and others. Mechanism by which these bioactive compounds exhibit a hypouricemic effect is summarized into three classes, namely the inhibition of uric acid production, improved renal uric acid elimination, and improved intestinal uric acid secretion [41]. Quercetin contained in *Plectranthus* has many properties. The results showed that quercetin decreased the increase in uric acid concentrations in healthy women [42].

CONCLUSION

The conclusion of this study was that the *Plectranthus amboinicus* L Spreng leaf meal supplementation significantly affected the performance of laying hens by decreasing weekly body weight, carcass weight and meat cholesterol significantly compared to controls. Feed PLP supplementation increases the percentage of cassava and feed conversion. PLP egg quality did not affect egg weight but significantly reduced egg yolk cholesterol. Feeding PLP supplementation significantly reduces uric acid levels in the blood.

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