

## THE EFFECT OF *Sauropus androgynus* LEAF EXTRACT ON PERFORMANCE, EGG QUALITY AND CHEMICAL COMPOSITION OF EGGS

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### ABSTRAK

Penelitian ini bertujuan untuk menghasilkan telur yang kaya nutrisi, tetapi rendah kolesterolnya. Empat puluh delapan ayam petelur strain Dekalb Warren umur 72 minggu dikelompokkan menjadi enam kelompok perlakuan. Satu kelompok, ayam petelur diberi pakan tanpa ekstrak daun katuk (*Sauropus androgynus*, EDK) sebagai kontrol, dan lima kelompok lainnya diberi pakan yang mengandung masing-masing 9 g EDK, 18 g EDK, 27 g EDK, 36 g EDK, dan 45 g EDK/kg pakan. Rancangan acak lengkap digunakan dalam penelitian ini. Hasil penelitian menunjukkan bahwa suplementasi EDK berpengaruh tidak nyata terhadap produksi telur, konsumsi pakan, konversi pakan, berat telur, berat kuning telur, albumen, berat badan, kulit telur, indeks telur, ketebalan kulit telur, dan indeks kuning telur ( $P > 0,05$ ), tetapi secara nyata berpengaruh terhadap Haugh Unit, indeks albumen, warna kuning, bau telur, rasa telur, dan warna kuning ( $P < 0,05$ ). Kadar protein, lemak, kalsium, fosfor, kalium dan besi dalam telur tidak berubah ( $P > 0,05$ ), namun kadar kolesterol telur secara nyata menurun ( $P < 0,05$ ). EDK meningkatkan kadar vitamin A dan  $\beta$ -karoten ( $P < 0,05$ ), dan memodifikasi komposisi asam lemak dan asam amino dalam telur. Dapat disimpulkan bahwa suplementasi EDK tidak meningkatkan performa, tetapi memperbaiki kualitas telur dan komposisi kimia telur.

*Kata kunci: ekstrak daun katuk, kualitas telur, performa, komposisi telur*

### ABSTRACT

The purpose of this research was to produce the enriched eggs, but low in cholesterol content. Forty eight layer chickens aged 72 weeks (strain Dekalb Warren) were distributed into six treatment groups. One group of laying hens was fed diets without *Sauropus androgynus* leaf extract (SALE) as the control, and another five groups were fed diet supplemented to 9 g SALE, 18 g SALE, 27 g SALE, 36 g SALE, and 45 g SALE/kg diet, respectively. Completely randomized design was used in the present study. The experimental results showed that SALE supplementation had no effect on egg production, feed intake, feed conversion ratio, egg weight, yolk weight, albumen weight, shell egg, egg index, eggshell thickness, and yolk index ( $P > 0.05$ ), but significantly affected Haugh Unit, albumen index, yolk color, egg odor, egg taste, and yolk color ( $P < 0.05$ ). The protein, fat, calcium, phosphorus, potassium and iron contents of eggs did not change ( $P > 0.05$ ), but the cholesterol content of egg was significantly reduced ( $P < 0.05$ ). In conclusion, the supplementation of SALE did not enhance performance, but it improve egg quality and its chemical composition.

*Keywords: Sauropus androgynus leaf extract, egg quality, performance, egg composition*

## INTRODUCTION

In the past, researchers emphasized their research to poultry nutrient requirements to support maximum performance. However, recently, they are very interested in enriching or altering the levels of certain nutrients in eggs. This trend is caused by the demands of consumers who want eggs that are rich in nutrients but low in cholesterol (Park *et al.*, 2005; Walker *et al.*, 2012).

Consumer demand for low-cholesterol eggs is very reasonable, because there is a positive correlation between consuming high-cholesterol eggs and an increase in the risk of atherosclerosis, which can result in up to a coronary heart attack, stroke or other metabolic diseases (Oh *et al.*, 2005). However, it is not easy to lower cholesterol levels without lowering egg production and egg weight (Santoso *et al.*, 2005; Yalcin *et al.*, 2007; Santoso *et al.*, 2010b). In addition, consumers also need eggs, which are enriched in  $\beta$ -carotene, protein and amino acid, unsaturated fatty acids, especially docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA), minerals and vitamins (Harris *et al.*, 2007; McCully, 2007; Omenn, 2007). All consumer demands can be met by modifying the nutrients through the addition of a feed additive. However, research on the enrichment of nutrients in eggs as far only focus on one nutrient alone (Park *et al.*, 2005). If the feed additive only provides one type of nutrient enrichment, this requires a lot of feed additive which would be difficult in practice. Thus, it is necessary to find a more effective feed additive, for enriching egg nutrients while lowering cholesterol without lowering production. The potential feed additive for such purposes is *Sauropus androgynus* leaf extract (SALE) (Santoso, 2001a,b,c), fish oil (Bovet *et al.*, 2007; Cherian, 2008) and vitamin E (Traber and Atkinson, 2007).

Samad *et al.* (2014) reported that the active compounds in *Sauropus androgynus* leaf are mainly fatty acid, chlorophyll, alkaloid, glycerol and benzoic acid. *Sauropus androgynus* leaf is rich in phenolics, flavonoid, and ascorbic acid, which plays an important role as an antioxidant (Supavanich *et al.*, 2012), glutamic acid (Santoso, 2014) and methylpyroglutamate (Agustal *et al.*, 1997), which plays as main compounds in the taste of poultry product,  $\beta$ -carotene and iron (Santoso *et al.*, 2015). Santoso *et al.* (2015) reported that the inclusion of *Sauropus*

*androgynus* leaf powder increased protein,  $\beta$ -carotene, vitamin A and iron contents of broiler meats.

The present research was conducted to evaluate the effect of *Sauropus androgynus* leaf extract on performance, egg quality and chemical composition of eggs.

## MATERIALS AND METHODS

### Extraction of *Sauropus androgynus* Leaf

*Sauropus androgynus* leaf was dried, and then ground into powder. The powder was then boiled at 60°C for 30 minutes in which the ratio *Sauropus androgynus* leaf with water was 1: 10. After that, the filtrate was filtered. Extraction was done twice. The SALE was then dried at 50°C for 36 hours.

### Maintenance of Laying Hens

This study used laying hens aged 72 weeks (strain Dekalb Warren). Completely randomized design was used in the present study. The diet contained 16.5% crude protein and 2,800 kcal ME/kg (NRC, 1994) without antibiotic supplementation.

Our previous results (Santoso *et al.*, 2005) showed that the supplementation of SALE at level of 9 g/kg diet was effective to reduce cholesterol content of egg without reducing egg production. It was assumed that extraction of the leaf at 90°C would impair active compounds of the leaf, so that it may be benefit if the temperature of extraction was lower than 90°C. The level of the leaf extract at previous results was used as basic supplementation of the leaf extract. Forty-eight laying hens were grouped into six treatment groups as follows: 1) Laying hens were fed diet without SALE as the control (P0); 2) Laying hens were fed diet with 9 g SALE/kg diet (P1); 3) Laying hens were fed diet with 18 g SALE/kg diet (P2); 4) Laying hens were fed diet with 27 g SALE/kg diet (P3); 5) Laying hens were fed diet with 36 g SALE/kg diet (P4); and 6) Laying hens were fed diet with 45 g SALE/kg diet (P5).

Eight laying hens for each group were maintained in individual cages. The composition of experimental diet is listed in Table 1. Laying hens were fed for a 30-day trial. They were fed diet for 120 g/day /bird. Feed intake, feed conversion ratio and egg production were measured weekly. Drinking water was given *ad libitum*.

Table 1. The Composition of Experimental Diets

Feedatuffs (g/kg)	0 g SALE	9 g SALE	18 g SALE	27 g SALE	36 g SALE	45 g SALE
Yellow corn	510	501	501	492	492	483
Soybean meal	140	140	140	140	140	140
Rice bran	200	200	191	191	182	182
Fish meal	70	70	70	70	70	70
SALE	0	9	18	27	36	45
Oil	10	10	10	10	10	10
Calcium carbonate	35	35	35	35	35	35
Mineral mixture	30	30	30	30	30	30
Premix	5	5	5	5	5	5

SALE = Sauropus androgynus Leaf Extract

### Sampling and Laboratory Analysis

A total of four eggs in each group were collected, and then analyzed the levels of protein, amino acids,  $\beta$ -carotene, fatty acids, calcium, phosphorus, kalium, iron, cholesterol and vitamin A in egg yolk. Protein content was determined by the method of AOAC (2012). Cholesterol content was determined spectrophotometrically at wavelengths of 490 nm by the method of Searcy and Berquist (1960) as modified by Bohac *et al.* (1988), while saponification was done by adding about 3% of pyrogalol (Živković *et al.*, 2002). Amino acid composition was measured by the method as described by Morel *et al.* (2003). The lipids were extracted according to Folch *et al.* (1957) with a chloroform-methanol mixture, and methylized by 20% boron trifluoride methanol complexin methanol solution (Morrison and Smith, 1964). Fatty acid composition was then determined by gas chromatography.  $\beta$ -carotene, iron, and vitamin A were measured by the method of Slamet *et al.* (1990) and Subekti (2003).

To test the quality of the eggs, the eggs were measured weight, eggshell thickness, Haugh Unit, air sac, yolk color and organoleptic tests. To test organoleptic, ten trained sensory panelists were asked to compare the relative palatability of taste and fishy odor. The yolk color test was done by comparing the color of the yolk with yolk color scale. The odor was assessed based on the value of 1 (very fishy), 2 (fishy), 3 (slightly fishy), 4 (less fishy) and 5 (not fishy). Panelists were also asked to taste and rate the taste of bad eggs (value

1) to very good (score 5). The eggs boiled at a temperature of 80 ° C for 20 minutes, cooled and tested the odor and taste.

### Data Analysis

The data of the study were analyzed by Anova and if significantly different then were tested further by Duncan's multiple range test.

## RESULTS AND DISCUSSION

### Performance of Laying Hens

The effect of the SALE on the performance of laying hens is presented in Table 2. The experimental results showed that the SALE did not significantly affect egg production, feed intake and feed conversion ( $P>0.05$ ). The results of this study differ from previous research results (Santoso *et al.*, 2005) who reported that egg production was increased by the supplementation of SALE at a level of 9 g/kg diet. This difference might be caused by differences in the manufacturing process of SALE where previously extracted at a temperature of 90°C while in the present study was extracted at a temperature of 60°C. Santoso *et al.* (2010) reported that supplementation of 90 mg non alkaloid extracted from *Sauropus androgynus* leaf also had no effect on egg production. Hermana *et al.* (2014) reported that feeding *Sauropus androgynus* leaf meal at a level of 10% had no effect on egg production of quails.

Table 2. The Effect of *Sauropus androgynus* Leaf Extract on Performance of Laying Hens

Variables	P0	P1	P2	P3	P4	P5
Egg production, egg/bird	21.4	22.3	22.1	23.8	20.8	22.1
Egg production, g/bird	1424.5	1485.0	1477.7	1539.6	1375.3	1454.6
Feed intake, g/bird	3594.9	3596.8	3598.4	3598.1	3599.1	3599.6
Feed conversion ratio	2.57	2.45	2.44	2.35	2.65	2.49

P0= Laying hens were fed diet without *Sauropus androgynus* leaf extract (SALE) as the control; P1= Laying hens were fed diet supplemented 9 g SALE/kg diet (P1); P2= Laying hens were fed diet supplemented 18 g SALE/kg diet; P3= Laying hens were fed diet supplemented 27 g SALE/kg diet; P4= Laying hens were fed diet supplemented 36 g SALE/kg diet; and P5= Laying hens were fed diet supplemented 45 g SALE/kg diet.

Table 3. The effect of *Sauropus androgynus* Leaf Extract on Fresh Egg Quality

Variables	P0	P1	P2	P3	P4	P5
Egg weight, g	66.57	67.39	66.97	64.98	66.25	65.90
Yolk weight, %	23.38	24.51	25.18	24.57	25.00	23.33
Albumen weight, %	63.83	62.65	62.04	63.64	62.41	64.26
Shell weight, %	12.80	12.84	12.78	11.80	12.59	12.41
Egg index	0.71	0.77	0.72	0.74	0.75	0.75
Shell thickness, mm	0.5	0.5	0.5	0.5	0.5	0.5
HU	68.24 <sup>a</sup>	59.82 <sup>a</sup>	82.83 <sup>b</sup>	64.03 <sup>a</sup>	70.96 <sup>a</sup>	84.11 <sup>b*</sup>
Albumen index	0.058 <sup>a</sup>	0.045 <sup>a</sup>	0.095 <sup>b</sup>	0.057 <sup>a</sup>	0.065 <sup>a</sup>	0.092 <sup>b*</sup>
Yolk index	0.418	0.430	0.455	0.439	0.423	0.439
Yolk color	5.62 <sup>a</sup>	6.93 <sup>b</sup>	7.56 <sup>b</sup>	8.0 <sup>bc</sup>	8.56 <sup>c</sup>	9.06 <sup>c**</sup>
Air sac, mm	2.61	3.14	2.76	2.77	3.31	2.72

Explanation of P0-P5: see Table 2

### Egg Quality

Table 3 shows the effect of *Sauropus androgynus* leaf extract on fresh egg quality. The results showed that administration of the SALE did not significantly affect egg weight, yolk weight, albumen weight, shell weight, egg index, eggshell thickness, the depth of the air sac, and yolk index ( $P>0.05$ ), but significantly affected Haugh Unit (HU), albumen index and yolk color ( $P<0.05$ ). P2 and P5 had higher HU and albumen index than the other treatment groups ( $P<0.05$ ). P0 had lower yolk color than the other treatment groups. *Sauropus androgynus* leaf is rich in  $\beta$ -carotene that may partly cause an increase in yolk

color. The results of this study differ from previous research where laying hens were given 9 g SALE/kg diet, which showed no increase in yolk color (Santoso *et al.*, 2005) and HU (Santoso, 2007). This difference is due to the different extraction methods and also due to the level of administration in this study was higher. Hermana *et al.* (2014), Subekti (2003) and Wiradimadja *et al.* (2010) reported that feeding *Sauropus androgynus* leaf meal increased egg yolk color. The yolk color enhancement may be caused by the presence of elevated levels of  $\beta$ -carotene in eggs (see Table 5). *Sauropus androgynus* leaf contains  $\beta$ -carotene at a level of

3,510.3 µg/g (Santoso, 2014; Santoso *et al.*, 2015).

Table 4 presents the effect of *Sauropus androgynus* leaf extract on organoleptic characteristics of eggs. The results showed that administration of SALE significantly reduce the odor, and improve the taste and yolk color ( $P < 0.05$ ). The odor of eggs significantly decreased in the presence of SALE. The present study was in agreement with the observation of Santoso (2007) who reported that the supplementation of SALE at level of 9 g/kg diet reduced fishy odor of eggs by 22.9%. Compounds that play a role in the decline of the odor is not yet known. Krishnaiah *et al.* (2007) stated that plants produce an antioxidant compound that includes carotenoids, flavonoids, cinnamic acids, benzoic acids, folic acid, vitamin C, Vitamin E and tocotrienols, vitamin A, palmitic acid,  $\beta$ -sitosterol, selenium,

anthraquinone, tannic acid. Katuk leaf was rich in  $\beta$  carotene, vitamin C, vitamin E and flavonoid (Andarwulan *et al.*, 2012), and other phenolic compounds (Nahak and Sahu, 2010), palmitic acid (Santoso, 2014) and chlorophyll (Samad *et al.*, 2014), protein (Madhu *et al.*, 2014). These compounds might contribute inhibiting oxidation of fatty acid of eggs, and therefore reduce the fishy odor of eggs.

An improve in egg taste might relate to glutamic acid content of eggs. Santoso (2014) stated that *Sauropus androgynus* leaf is rich in glutamic acid, whereas Agustal *et al.* (1997) reported that *Sauropus androgynus* leaf is rich in methylpyroglutamate, which may be converted into glutamic acid in the gastrointestinal tract. This study showed that to enhance the taste of eggs, the extract should be supplemented at least at level of 36 g/kg diet. Santoso (2007) also

Table 4. The Effect of *Sauropus androgynus* Leaf Extract on Organoleptic Characteristics of Eggs

Variables	P0	P1	P2	P3	P4	P5
Whole egg odor	2.4 <sup>b</sup>	2.3 <sup>b</sup>	2.4 <sup>b</sup>	1.7 <sup>ab</sup>	1.30 <sup>a</sup>	1.45 <sup>a</sup>
Egg odor after peeling	3.6 <sup>b</sup>	3.4 <sup>b</sup>	3.2 <sup>b</sup>	2.45 <sup>a</sup>	2.30 <sup>a</sup>	2.00 <sup>a</sup>
Egg odor after split	3.45 <sup>b</sup>	3.05 <sup>b</sup>	3.05 <sup>b</sup>	2.70 <sup>ab</sup>	2.30 <sup>a</sup>	2.25 <sup>a</sup>
Egg taste	2.50 <sup>a</sup>	2.95 <sup>a</sup>	3.05 <sup>a</sup>	3.15 <sup>a</sup>	3.75 <sup>b</sup>	4.35 <sup>b*</sup>
Yolk color	1.88 <sup>a</sup>	2.06 <sup>a</sup>	2.94 <sup>b</sup>	4.00 <sup>c</sup>	4.38 <sup>c</sup>	4.81 <sup>c**</sup>

Explanation of P0-P5: see Table 2

Table 5. The Effect of *Sauropus androgynus* Leaf Extract oComposition of Egg

Variables	P0	P1	P2	P3	P4	P5
Protein, %	16.60	16.52	16.63	16.69	16.76	16.69
Fat, %	31.90	31.85	31.89	31.85	31.86	31.86
Calcium, mg/l	147.08	147.12	147.14	147.11	147.09	147.13
Phosphor, mg/l	586.04	586.10	586.04	586.07	586.10	586.06
Iron, mg/l	7.25	7.23	7.27	7.25	7.28	7.28
Kalium, mg/l	134.32	135.30	136.21	136.62	137.24	137.55
Cholesterol, mg/100 mg	3.03 <sup>b</sup>	2.74 <sup>ab</sup>	2.55 <sup>a</sup>	2.53 <sup>a</sup>	2.41 <sup>a</sup>	2.31 <sup>a*</sup>
Vitamin A (IU)	1986.9 <sup>a</sup>	2278.9 <sup>a</sup>	2271.5 <sup>a</sup>	2542.1 <sup>b</sup>	2523.5 <sup>b</sup>	2754.0 <sup>b**</sup>
$\beta$ -carotene (µg/100g)	632.0 <sup>a</sup>	662.8 <sup>a</sup>	761.5 <sup>b</sup>	800.9 <sup>b</sup>	816.0 <sup>b</sup>	893.4 <sup>b*</sup>

Explanation of P0-P5: see Table 2

reported that supplementation of this extract at level of 9 g/kg diet did not improve the taste of eggs.

### Egg Composition

Table 5 shows the effect of *Sauropus androgynus* leaf extract on composition of egg. The results showed that the SALE supplementation did not significantly affect ( $P>0.05$ ) the levels of protein and fat in the yolk. This indicates that SALE was unable to lower the fat content of eggs. There are mechanisms in the chicken's body to retain fat content in eggs for normal reproductive function. That is why the levels of fat in eggs are difficult to be lowered. The results of this study are different from when SALE was given in broilers where both levels of these nutrients can be modified (Santoso *et al.*, 2013).

The results showed that the SALE supplementation did not significantly affect levels of calcium, phosphorus, iron, and kalium, but significantly reduced levels of cholesterol ( $P<0.05$ ), and increased ( $P<0.05$ ) the levels of vitamin A and  $\beta$ -carotene in egg yolk. The P2, P3, P4, and P5 had lower cholesterol contents than the P0 ( $P<0.05$ ). The P3, P4, and P5 had higher vitamin A than P0, P1 and P2 ( $P<0.05$ ). The P2, P3, P4, and P5 had higher  $\beta$ -carotene than P0 and P1 ( $P<0.05$ ).

*Sauropus androgynus* leaf contains compounds that inhibit the metabolism of minerals. In addition, the availability of mineral

derived from plants is low. Thus, although *Sauropus androgynus* leaf is rich in mineral, it was unable to increase the content of the mineral in eggs.

The results of this study were similar to previous research results (Santoso *et al.*, 2005; Subekti *et al.*, 2006) who found that supplementation of SALE can lower cholesterol in egg yolk. Active compounds, which plays as anticholesterol include alkaloid (Santoso *et al.*, 2010b), phytosterol (Subekti, 2007) and flavonoid (Lien *et al.*, 2007).

An Increase in levels of vitamin A and  $\beta$ -carotene in egg yolk may be caused by  $\beta$ -carotene in *Sauropus androgynus* leaf. Subekti (2003) Hermana *et al.* (2014) and Wiradimadja *et al.* (2010) reported that feeding *Sauropus androgynus* leaf meal increased the content of vitamin A of egg yolk. The laboratory analysis in the present study showed that the SALE contains  $\beta$ -carotene as much as 3,328 mg/100g extract. Subekti (2003) reported that feeding *Sauropus androgynus* leaf meal increased the content of  $\beta$ -carotene of native chicken egg yolk. Santoso *et al.* (2015) reported that feeding fermented *Sauropus androgynus* leaf meal increased the content of  $\beta$ -carotene of broiler meats.  $\beta$ -carotene is converted to retinal in the intestinal mucosa, then to retinol (vitamin A). The key step for vitamin A formation is the oxidative cleavage of provitamin A carotenoids by  $\beta$ -carotene 15,15-dioxygenase (Thurnham, 2007).

Table 6. The Effect of *Sauropus androgynus* Leaf Extract on Yolk Fatty Acid Composition

Fatty acids, %	P0	P1	P2	P3	P4	P5
Lauric acid	0.008	0.011	0.008	0.039	0.004	0.005
Myristic acid	0.887	0.381 <sup>a</sup>	0.544 <sup>b</sup>	0.520 <sup>b</sup>	0.645 <sup>b</sup>	0.807 <sup>b*</sup>
Palmitic acid	27.354	23.235	24.366	26.226	25.131	18.692
Stearic acid	0.372 <sup>b</sup>	0.214 <sup>a</sup>	0.179 <sup>a</sup>	0.468 <sup>b</sup>	0.317 <sup>b</sup>	0.240 <sup>a*</sup>
Oleic acid	60.570	61.391	61.138	57.102	61.755	60.660
Linoleic acid	6.534	8.254	8.870	7.085	7.990	6.698
Saturated fatty acid	28.621	23.841	25.097	27.253	26.097	19.744
Unsaturated fatty acid	67.104	69.645	70.008	64.187	69.745	67.358
Unidentified fatty acid	4.275 <sup>a</sup>	6.514 <sup>a</sup>	4.895 <sup>a</sup>	8.560 <sup>b</sup>	4.158 <sup>a</sup>	12.898 <sup>c</sup>

Explanation of P0-P5: see Table 2

### Fatty Acid Composition

Table 6 shows the effect of *Sauropus androgynus* leaf extract on yolk fatty acid composition. SALE supplementation significantly reduced stearic acid ( $P < 0.05$ ). The supplementation of the extract as much as 9 g, 18 g or 45 g/kg diet reduced the levels of stearic acid when compared with the control. SALE supplementation also significantly reduced myristic acid ( $P < 0.05$ ), i.e. at the level of extract 9 g, 18 g or 27 g/kg diet. Santoso *et al.* (2015) reported that *Sauropus androgynus* leaves had myristic acid 8.81%, palmitic acid 48.73%, stearic acid 3.08%, oleic acid 6.72%, linoleic acid 5.11%, which mean that *Sauropus androgynus* is rich in saturated fatty acid than unsaturated fatty acid. It appears that supplementation of this extract reduced the digestibility and/or absorption of stearic acid and miristic acid. This study differ from the observation of Santoso *et al.* (2010a)

who reported that inclusion of SALE increased miristic acid of broiler meats. Another interesting result is the presence of unidentified compounds. These compounds apparently increased by supplementation of SALE.

### Amino Acid Composition

Table 7 presents the effect of *Sauropus androgynus* leaf extract on yolk amino acid composition. SALE supplementation significantly affected glutamic acid, arginine and lysine ( $P < 0.05$ ), but did not affect other amino acid. The P2 and P3 had higher glutamic acid than the P0 ( $P < 0.05$ ). The P4 and P5 had higher arginine than the P0 and P1 ( $P < 0.05$ ). The P4 and P5 had higher lysine than the P0 and P1.

Santoso *et al.* (2015) reported that *Sauropus androgynus* leaf is rich in glutamic acid, whereas Agustal *et al.* (1997) reported that *Sauropus androgynus* leaf is rich in methylpyroglutamate,

Table 7. The Effect of *Sauropus androgynus* Leaf Extract on Yolk Amino Acid Composition

Amino acids, %	P0	P1	P2	P3	P4	P5
Aspartic acid	1.660	1.493	1.902	1.386	1.679	1.742
Glutamic acid	2.278 <sup>a</sup>	2.312 <sup>a</sup>	2.757 <sup>b</sup>	2.816 <sup>b</sup>	2.324 <sup>a</sup>	2.420 <sup>a*</sup>
Serine	0.891	0.880	0.949	1.041	1.103	1.026
Glycine	0.370	0.299	0.377	0.320	0.386	0.456
Histidine	0.353	0.402	0.292	0.267	0.337	0.325
Arginine	0.574 <sup>a</sup>	0.646 <sup>a</sup>	0.748 <sup>ab</sup>	0.761 <sup>ab</sup>	0.844 <sup>b</sup>	1.178 <sup>c*</sup>
Threonine	0.505	0.455	0.544	0.553	0.589	0.494
Alanine	1.204	0.947	1.026	0.835	0.940	1.255
Proline	0.360	0.288	0.470	0.456	0.506	0.421
Tyrosine	0.520	0.466	0.489	0.446	0.603	0.583
Valine	1.356	1.342	1.378	1.044	0.994	1.133
Methionine	0.557	0.449	0.402	0.332	0.456	0.517
Cystine	0.345	0.451	0.280	0.333	0.437	0.330
Isoleucine	0.689	0.773	0.606	0.609	0.934	0.733
Leucine	1.444	1.818	1.845	1.223	1.709	1.345
Phenylalanine	0.501	0.602	0.613	0.406	0.647	0.482
Lycine	0.686 <sup>a</sup>	0.757 <sup>a</sup>	0.839 <sup>ab</sup>	0.882 <sup>ab</sup>	0.905 <sup>b</sup>	1.055 <sup>b</sup>
Sulfuric amino acid	2.346	2.718	2.527	1.888	2.602	2.192
Total amino acid	14.293	14.380	15.517	13.990	15.393	15.495

Explanation of P0-P5: see Table 2

which may be converted into glutamic acid. An increase in glutamic acid might stimulate arginine synthesis by bacteria, since Hood and Lyman (1950) reported that glutamic acid have an important role in arginine synthesis. In addition, there was also a small contribution of arginine from the extract. The mechanism of an increase in lysine is still unknown. It has been established that lysine is essential amino acid in animal, so that it is not synthesized in the body. Most bacteria synthesize lysine from aspartic acid. SALE increased the number of *Bacillus subtilis* (Santoso *et al.*, 2001) and *Lactobacillus sp* (Santoso *et al.*, 2001; Santoso, 2005). It is assumed that these bacteria synthesize lysine from aspartic acid. The SALE supplementation changes the composition of amino acids of the egg yolk, although the egg yolk protein levels unchanged. Santoso *et al.* (2015) reported that supplementation of *Sauropus androgynus* leaf meal changed the composition of amino acid of broiler meats.

## CONCLUSION

*Sauropus androgynus* leaf extract supplementation did not enhance the performance of laying hens, but improved egg quality and organoleptic properties. This extract also increased the contents of vitamin A and  $\beta$ -carotene, but reduced the content of cholesterol in eggs. However, it was unable to modify the content of protein, minerals and fat in the eggs.

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