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Effect of Spirulina (*Spirulina platensis*) on Internal Quality of Eggs in Laying Hens

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ABSTRACT

The research aims to determine the effect of adding Spirulina (*Spirulina platensis*) on egg yolk index, albumin index, egg yolk colour score, and *haugh unit*. The research were used 150 laying hens aged 89 weeks strain Hyline Brown were used in this study reared in 5 treatment, such as (P0) using 0% of *Spirulina platensis*, (T1) using 0,25% *Spirulina platensis*, (T2) using 0,50% *Spirulina platensis*, (T3) using 0,75% *Spirulina platensis*, and (T4) using 1,00% *Spirulina platensis*. In this study, Completely Randomized Design (CDR) were used for research method with five level of treatments and five replications. Each replicant used six laying hens. The observed variables were egg yolk index, albumin index, egg yolk colour score, and *haugh unit* value. If there were significantly different results, it will be continued with Duncan's Multiple Range Test (DMRT). The final result showed that Spirulina (*Spirulina platensis*) addition to layers feed didn't gave a significant effect ($P>0.05$) on egg yolk index, albumen index, and *haugh unit* value, but had a highly significant effect ($P<0.01$) on egg yolk colour score. The results of DMRT showed that the egg yolk colour at T4 replication was significantly higher than T0, T1, T2, and T3. The results of this study concluded that Spirulina (*Spirulina platensis*) with 1% level, can be applied to layers feed without having any negative effect on other internal egg quality.

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Introduction

Eggs are food ingredients derived from poultry with a fairly high nutritional value. In addition to having protein content, eggs also contain vitamins, minerals and fatty acids [1]. Eggs are also a source of animal protein with an excess of amino acids which are more complete than other sources of protein such as fish, tempeh, tofu and others [2]. Therefore, besides egg production, another thing that is considered is internal quality. The internal quality of the egg itself consists of the egg whites index, the yolk index, the *haugh unit*, and the color of the yolk [3]; [4]. Some of the factors that affect the internal quality of eggs are the life of the mother and also the length of storage of the

eggs. The cause of the decline in the internal quality of eggs during storage is due to evaporated egg components and also the transfer of water from egg whites to yolks [5]. The quality of eggs can be affected by the feed consumed by chickens. Nutrition in feed plays an important role in improving the quantity and quality of feed. Improving feed quality can be done by adding additive feed ingredients.

Spirulina (*Spirulina platensis*) is an edible microalga known for being highly nutritious and a potential feed source for various animal species [6]. Spirulina is a green-blue algae that can be found in brackish water that has alkaline properties [7]. Spirulina is known to help reduce heat stress in laying hens. Spirulina plays a role in improving animal welfare and stabilizing egg production in hot environmental conditions [6]. Selim, *et al.* [8] reported that the nutritional composition of Spirulina includes crude protein of 51.7% BK, crude fat of 9% BK, crude fiber of 4.5% BK, and ash content of 10.4% BK. The nutritional content of spirulina is rich in xanthophyll content which is also one of the ingredients in egg yolks that gives color to the yolk [9]. Spirulina can be used as an addition to chicken feed which aims to improve the quality of the internal chicken eggs. This is because the color of the yolk increases significantly due to the addition of Spirulina to the feed of laying hens due to pigmentation and also the high deposition of carotene derived from spirulina [10]. The addition of Spirulina as an additive feed ingredient can improve productivity, quality, egg shelf life, and egg health, thus allowing for more sustainable and efficient production of laying hens [11].

State of the Art

Laying Hens

Laying hens are livestock commodities that contribute to meeting animal protein needs through highly nutritious products [12]. Poultry farming is essential to meet the global demand for animal protein, and improving the health and efficiency of laying hens is essential for sustainability [11]. The laying hen farming industry is currently facing a dual challenge in the form of the challenge of meeting the increasing demand for animal protein and adapting to consumer preferences for superior quality eggs. The assessment of egg quality aspects is not only limited to external appearance (shape, strength, and color of the opener) but is also assessed from internal aspects which include the quality of egg yolk color, albumen height, yolk height, and *haugh unit* [12]. Efforts to meet this challenge are carried out by tracing additive feed ingredients that can improve the internal quality of eggs.

Spirulina platensis

Spirulina platensis with its bioactive compound content has emerged as a promising candidate for additive feed ingredients. Spirulina has a complete content of essential

amino acids, vitamins, minerals as well as dye compounds and bioactive compounds in it [13]. The concentration of keratinoids and natural dyes in *Spirulina* offers potential as an egg yolk coloring agent [11]. *Spirulina* is a natural product that is highly nutritious and contains antioxidant compounds with the ability to increase the production ability of both growth, hatching, and egg production [14].

Perception

Efforts to handle egg quality can be done by using certain additive feed ingredients. The use of *Spirulina platensis* as an additive feed ingredient is expected to have a biological mechanism that helps improve egg quality. [15] The addition of spirulina to the drinking water of laying hens can increase the color of the yolk, the antioxidant capacity of the yolk, and the increase of lipoproteins in the blood, and create potential profits for the laying hen farming business. The use of spirulina in laying hen feed close to the retired period at the level of 0.75% can reduce feed conversion [6]. The addition of spirulina was reported to have no adverse effects on egg production performance and egg quality characteristics and can be safely used in laying hen feed, at a rate of 0.3% with a beneficial effect on the yolk of aged laying hens [16]. The use of *Spirulina* in laying hen feed shows its capacity to provide added value to products in the form of superior internal quality of eggs with a more attractive yolk color for consumers. Until now, there are still opportunities and gaps to conduct research related to optimal doses and effective administration duration, a research is still needed. Thus *Spirulina* can become a component in the production of high-quality eggs and meet the demands of the modern market.

Method

Research Design

This study used a completely randomized design (CRD) using 5 treatments and 5 replications. The treatment consists of 5 treatments, there are:

T0 : Control feed

T1 : Feed with 0,25% *Spirulina plantesis*

T2 : Feed with 0,50% *Spirulina plantesis*

T3 : Feed with 0,75% *Spirulina plantesis*

T4 : Feed with 1,00% *Spirulina plantesis*

Cage Preparation

The cage used in this study are open house system cages that have feeders and drinkers (nipple drinkers). The cages used are battery-shaped which consists of 3 levels and uses 75 batteries, each cage is filled with 2 egg-laying hens, each bulkhead has 6 egg-laying hens so that the number of egg-laying hens used in this study

amounted to 150 egg-laying hens. The laying hens used in this study were 89 weeks old.

Research Preparation and Feeding Management

Preparation was carried out with a formulation for the standard needs of layer phase. The nutritional content of the basal feed used is moisture content of 13%, PK 17%, LK 3%, SK 7%, ash 14%, calcium 3.25-4.25%, phosphorus 0.45%, along with amino acids lysine 0.80%, methionine 0.40%, methionine + cystine 0.67%, tryptophan 0.18%, and threonine 0.55%. The composition of basal diet is corn with a percentage of 50%, concentrate 35%, and rice bran 15%. The process of mixing the ration was done manually, and then the feed was served in crumble form. Each cage was fed twice a day, in the morning and evening with 120g/head/day feeding. Drinking water was provided ad libitum during data collection. The composition of the feed was shown in Table 1.

Research Variables

Data was collected every day from week 89th of age to week 95th of age with the following parameters:

Yolk Index

The yolk index can be used to determine the viscosity of the yolk.. Yolk index measurement is carried out using the yolk height and yolk diameter components [17]. The formulation used is as follows.

$$Yolk\ Index = \frac{Yolk\ Height\ (mm)}{Yolk\ Diameters\ (mm)}$$

Albumen Index

The albumen index is the ratio of the height of thick albumen to its average diameter [18]. The formulation used to measure the albumen index is as follows.

$$Albumin\ index = \frac{Ta}{(Da + Db):2}$$

Description:

Ta: Albumin Height

Da: Longest diameter of albumin

Db: Shortest diameter of albumin

Yolk Color Score

The color of egg yolk is one of the variables of the internal quality of egg yolk. For the color score on the yolk itself using the Egg Yolk Color Fan tool which has a colour scale of 1-15. The higher the value, the better

Haugh Unit

Haugh unit is the quality of albumin measured by egg weight and albumin height. The higher haugh unit value means that the egg has better quality [19]. The formula of haugh unit is expressed as follows.

$$\text{Haugh unit (HU)} = 100 \log (H + 7,57 - 1,7 W^{0,37})$$

Description:

H = Albumin height (mm)

W = Egg Weight (gram)

Data Analysis

Data was analyzed using the Analysis of Variance (ANOVA) test using the Microsoft Excel. Treatment was considered significant if the P value <0.05. Duncan's Multiple Range Test (DMRT) was performed to detect differences between treatments.

Results and Discussion

Data from the research showed information the effect of the addition *Spirulina platensis* to the ration of laying hens on the internal quality of chicken eggs which can be seen in Table 1. Based on the results data analysis, it was found that the addition of *Spirulina platensis* did not significantly affect ($P>0.05$) the internal quality of eggs in the form of yolk index, albumen index, yolk colour score and haugh unit. However, it gave a very significant effect ($P<0.01$) on the yolk colour score.

Table 1. Results of Analysis of Egg Yolk Index, Egg White Index, Egg Yolk Colour Score, and Haugh units.

Treatments	Variable			
	Yolk Index	Albumin Index	Yolk Color Score	Haugh unit
T0	0,4105±0,0162	0,071±0,011	9,37±0,69 ^a	71,8199±6,584
T1	0,4094±0,0193	0,071±0,012	9,74±0,23 ^a	71,8341±6,177
T2	0,4159±0,0105	0,068±0,005	10,06±0,52 ^{ab}	71,2881±2,200
T3	0,4109±0,0156	0,067±0,006	10,29±0,49 ^b	69,8030±1,378
T4	0,4194±0,0148	0,068±0,003	10,65±0,22 ^b	69,8331±2,507

Description:

Superscript ab in the same column indicates a very significant difference ($P < 0.01$); T0 = (control); T1 = PB + *Spirulina platensis* 0.25%; T2 = PB + *Spirulina platensis* 0.50%; T3 = PB + *Spirulina platensis* 0.75%; and T4 = PB + *Spirulina platensis* 1.00%.

Yolk Index

The results of the analysis variance of the research that studied the effect of spirulina on egg yolk index can be seen in Table 1, obtained the average value of egg white index with a trend from the highest and lowest in a row were T4 (0.4194 ± 0.0148), T2 (0.4159 ± 0.0105), T3 (0.4109 ± 0.0156), T0 (0.4105 ± 0.0162), and T1 (0.4094 ± 0.0193). The results of the study of the effect of *Spirulina platensis* on the yolk index can be seen in Figure 1. Based on Table 1 proves that the treatment of *Spirulina platensis* given to the laying hen feed with a percentage of 0.25%, 0.50%, 0.75%, and 1% does not have a significant effect ($P > 0.05$).

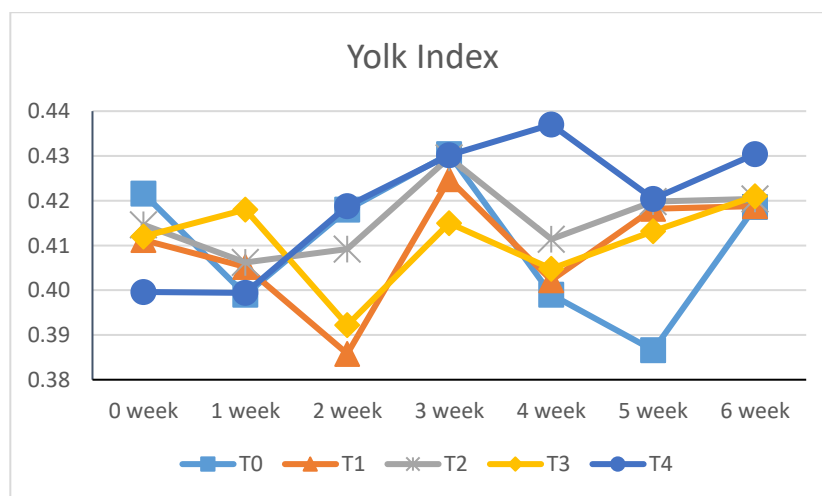


Figure 1. Yolk index graph of each treatment every week

There are several things that affect the yolk index, according to [20] stated that storage and storage temperature affect the yolk index. It was caused by the decreasing vitelin membrane and the thinner yolk which causes the diffusion of water from the egg white to the yolk, this is the cause of the enlarged yolk diameter and the decreasing elasticity of the vitelin membrane, the vitelin membrane will weaken its strength if the egg storage time is longer, this was caused by microorganisms entering the egg white and creating proteolytic enzymes and making the vitelin membrane weaken [21]. For the yolk index in this study, it has got a good value [22] and states that a good yolk index has a value between 0.33 and 0.50 and the average value was 0.42 [23].

In Figure 1 it can be seen at the beginning of the study that the yolk index in T4 was the lowest of all treatments. However, after 6 weeks it can be seen that the increase in yolk index in T4 was the greatest of all treatments, although when tested statistically at the end of the study there was no difference. The addition of spirulina in layer feed tended to increase the highest yolk index in T4, followed by T3, T2 and T1, respectively.

Albumen Index

The results of the variety-based analysis of the effect of spirulina on the albumen index can be seen in Table 1, the average egg white index value with the tendency of the highest and lowest in succession were T1 (0.071 ± 0.012), T0 (0.071 ± 0.011), T4 (0.068 ± 0.003), T2 (0.068 ± 0.005), and T3 (0.067 ± 0.006). The treatment of giving *Spirulina platensis* to laying breed chicken feed with percentages of 0.25%, 0.50%, 0.75%, and 1% did not have a noticeable effect ($P > 0.05$). There was no difference in the albumen index value is suspected because there is no difference in the protein content of the feed. Based on previous research, conducted [8] which stated that there was no significant difference between the control treatment and the feed added by *Spirulina platensis* to the albumen index. In their study, with the addition of 0.1%, 0.2%, and 0.3% of *Spirulina platensis*, the albumen index results ranged from 9.93%-14.7%.

Feed that has a high protein content will contribute protein to albumen in high amounts [24]. Methionine becomes an amino acid that affects the formation of the structure of albumen and exerts an influence on the meshes of ovomucin. Ovomucin has a role in creating a gel structure in albumin, the more and stronger the ovomucin meshes, the more viscous the albumin will become thicker and make the viscosity of albumin higher. The high content of ovomucin will make the egg whites able to maintain the freshness and thickness of albumin well.

The average results of the albumen index during the study in Table 1. Provide the albumen index value given *Spirulina platensis* feed supplement is at a reasonable value of 0.067-0.071, where according to Standart National Indonesia [25], the value of fresh albumen index ranges from 0.050-0.174. Based on a statement from which states that the longer the storage of eggs will make the value of the egg white index decrease, this was due to the evaporation of water and also CO₂ gas which makes the egg whites that were initially thick become liquid [26].

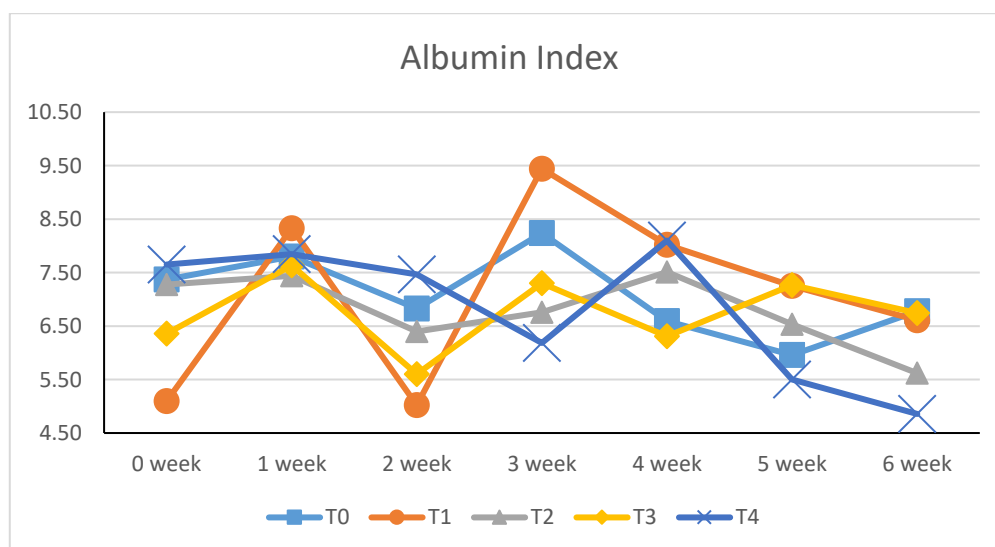


Figure 2. Albumen index graph for each treatment each week

In Figure 2, it can be seen that the value of each treatment is quite variable. Where in T1 the most extreme increase and decrease in value. On the first day of the study, T4 had the highest albumen index value, but on the last day of the research, T4 had the lowest value compared to the other treatments.

Egg yolk color score

The results of the variegated analysis of the effect of spirulina on the yolk color score can be seen in Table 1. It can be seen that the highest and lowest values are T4 (10.65), T3 (10.29), T2 (10.06), T1 (9.74), and T0 (9.37). The treatment of giving *Spirulina platensis* was carried out on laying breed chicken feed with a percentage of 0.25%, 0.50%, 0.75%, and 1% to have a very real effect ($P < 0.01$). [27] Argue that one of the indicators used to determine egg quality is the color of the yolk. According to research that has been conducted by [28] which found that the addition of *Spirulina platensis* to laying hen feed gave very real results. Obtained the following results for the egg yolk color score, 10.55 (Spirulina 1.5%), 11.43 (Spirulina 2.0%), and 10.66 (Spirulina 2.5%) while eggs from laying hens that ate control feed only got a yolk color score of 1.55 (basal feed). Stated that the influence of spirulina on the egg yolk color was more due to the content of carotenoid pigments possessed by *Spirulina platensis* [29]. Argue that the impact of the use of Spirulina on the color of the yolk was caused by a dose with an increased level of inclusivity as well as showing a clearer increase in the color of the yolk [11]. *Spirulina platensis* contains a type of bioactive compound that is a group of carotenoids, namely beta carotene. Beta carotene is an organic pigment that has an orange, red, orange, or yellow color that is created naturally in plants that carry out photosynthesis, algae, and certain types of fungi and bacteria [30]. Livestock that consume more

carotenoid pigments will give an egg yolk color with high intensity. The pigment that gives the yolk color will be absorbed by the small intestine which will then be distributed to the target organ in need [31]. The egg yolk color score obtained from this study was quite good, ranging from 9.37-10.61, because stated that people like the egg yolk color which has a score between 9-12 [32].

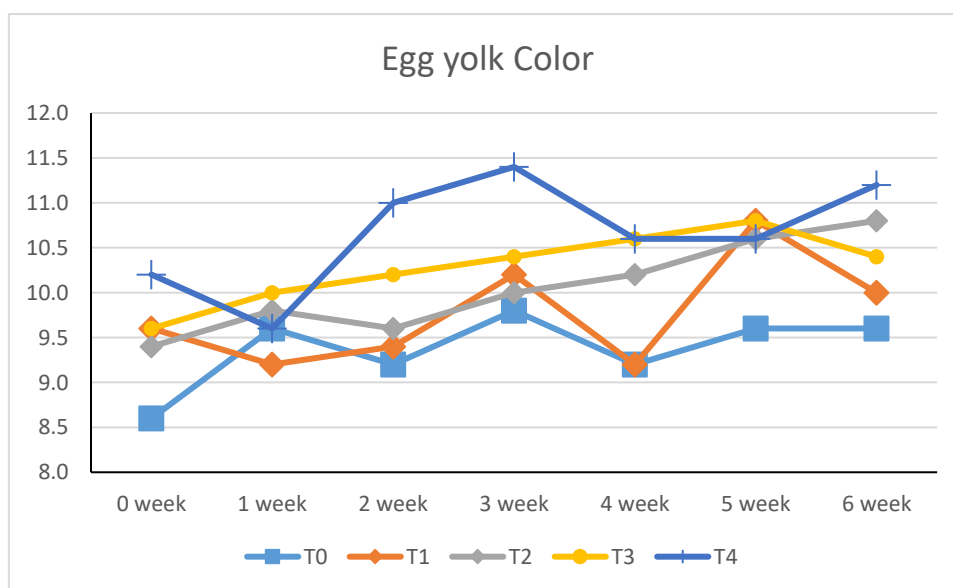


Figure 3. Egg yolk score graph for each treatment each week

In Figure 3, it is known that the egg yolk score at T4 has the greatest value, but it had decreased in the first week but increased and peaked in the 3rd week and in week 6 had the highest egg yolk score in each treatment. From the graph obtained, it can be seen that treatment 1 to 4 continues to experience an upward trend.

Haugh Unit

The results of the various analyses of the effect of spirulina on the *unit haugh* value can be seen in Table 1, the average *unit Haugh* value in chicken eggs is obtained with the highest and lowest respectively T1 (71.8341), T0 (71.8199), T2 (P1.2881), t4 (69.8331) and T3 (69.8030). The treatment of giving *Spirulina platensis* was carried out on laying breed chicken feed with percentages of 0.25%, 0.50%, 0.75%, and 1% did not have a real effect ($P > 0.05$). The *unit haugh value* was the accepted unit for measuring the albumin quality of an egg. Of all the types of egg quality, only the *haugh unit* is recognized as the best mathematical indicator of egg quality [33]. The *Haugh unit* itself comes from a calculation involving the height of the thick egg white with the weight of the egg. The smaller the *haugh value of the unit*, the lower the quality of the eggs will

be. The results of the research obtained are comparable to previous research conducted by [34] which found that there was no difference in the *haugh unit* value after the addition of *Spirulina platensis* with various concentrations. The results they got were 89.33 (0% spirulina supplement), 87.16% (5% spirulina supplement), 87.84% (10% spirulina supplement), and 90.08% (15% spirulina supplement).

Stated that the value of the *haugh unit* is influenced by the storage time and temperature [35]. Stated that eggs that are stored for longer will create a decrease in the *haugh unit* value, this can happen because CO₂ gas evaporates and makes the thick egg whites become diluted [36]: [37] also stated that the storage temperature can affect the egg haugh unit, the high storage temperature will encourage the rupture of ovomucin-lysozyme which makes the haugh unit of eggs stored at room temperature significantly reduced compared to those stored in the refrigerator. The haugh value obtained is quite good, because it has a value that ranges from 69.8030-71.8341 where the haugh unit value that is between 60-72 will get an A grade.

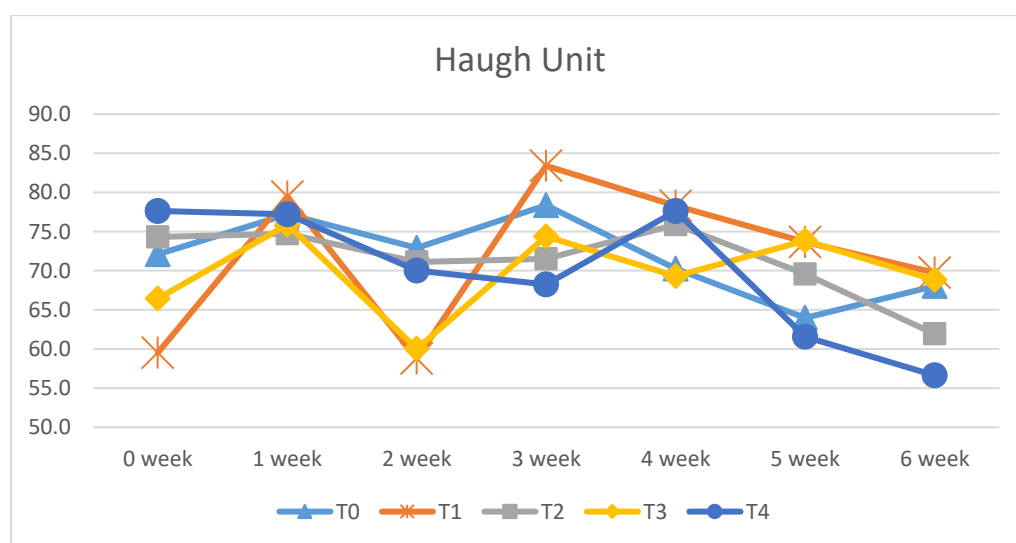


Figure 4. Graph of the haugh unit value of treatment units per week

In Figure 4, it can be seen that T4 in the first week had the highest haugh unit value compared to other treatments, but on the last day of the study the value decreased to the lowest. On the graph, the *haugh value* of this unit also experiences fluctuating values. Of all the treatments, only T1 increased at the end of the research when compared to the haugh unit value on first day research.

Conclusions

Based on the results of this study, a conclusion can be made that the administration of *Spirulina platensis* was able to improve the color of the yolk, but does not affect the white index, yolk index and *haugh unit*. It is recommended to use 1 % spirulina in feed to improve the color of the yolk. As well as using spirulina and combining with other additives to improve the quality of other eggs and egg production.

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