

Physical Quality of Eggs in Pengging Duck (*Anas platyrinchos*) after Administration of Nanochitosan as a Feed Additive

SUNARNO*, NURUL ANNISA* & SRI ISDADIYANTO

Department of Biology, Faculty of Science and Mathematics, Diponegoro University, Jalan Prof. Soedharto SH, Tembalang - Semarang 50275, Indonesia

*Corresponding authors: sunarno@lecturer.undip.ac.id; nurul.a459@gmail.com

Received: 21 December 2022

Accepted: 22 May 2023

Published: 30 June 2023

ABSTRACT

The problem often encountered by laying duck breeders is a decrease in the physical quality of eggs. Nanochitosan is a polysaccharide that has the potential as a feed additive to improve the physical quality of eggs in pengging ducks. The use of nanochitosan at levels of 2.5, 5.0, 7.5 and 10.0 g/kg of feed can improve digestive performance and nutrient absorption in ducks. The purpose of this study was to determine the effect of nanochitosan as a feed additive on the physical quality of eggs in pengging ducks. This study used a completely randomised design with five groups and five ducks per group. The treatments consist of control and the addition of nanochitosan as a feed additive with a concentration of 2.5, 5.0, 7.5 and 10.0 g/kg of feed. The variables observed in this study included the haugh unit (HU), yolk index (IKT), yolk weight, white weight, egg weight, shell thickness, and egg shape index. The research data were analysed using ANOVA with a significance level of 5%. The results showed that the addition of nanochitosan at different levels (2.5, 5.0, 7.5 and 10.0 g/kg of feed) had a significant effect on the measured egg quality consisting of HU, IKT, egg yolk weight, egg white weight, egg weight, egg, shell thickness, and egg shape index. The study concluded that nanochitosan can be used as a feed additive to improve the physical quality of eggs in pengging ducks. The use of nanochitosan with different levels as a feed additive can be useful for others by providing a reference about the levels of nanochitosan used as a feed additive which can improve the physical quality of duck eggs and become important information in increasing exploration in the use of nanochitosan products. This study is expected to open opportunities for further research in the future about the mechanistic effects of nanochitosan in improving egg quality and productivity of laying ducks.

Keywords: Egg quality, feed additives, nanochitosan, pengging duck

Copyright: This is an open access article distributed under the terms of the CC-BY-NC-SA (Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International License) which permits unrestricted use, distribution, and reproduction in any medium, for non-commercial purposes, provided the original work of the author(s) is properly cited.

INTRODUCTION

Duck farming has great potential to be developed in fulfilling community animal protein. Pengging ducks are a type of local duck originating from the Boyolali area, Central Java, Indonesia (Kasiyati *et al.*, 2021). The advantage of Pengging ducks is that they have a high level of productivity and the ability to adapt to a good environment (Kasiyati *et al.*, 2019). This type of local duck has a production period of 9 – 11 months per year (Wulandari *et al.*, 2015). Pengging ducks are superior to laying ducks because they can produce more than 200 eggs per year. The egg production of this poultry is more when compared to other local ducks such as Tegal ducks and Magelang ducks (Gibran *et al.*, 2021).

The problem that is often found by duck egg breeders is a decrease in the physical quality of

eggs. The great egg quality can be determined based on physical indicators of eggs which include egg weight, egg white weight, yolk weight, shell thickness, haugh unit (HU), yolk index (IKT), and egg shape index (Saraswati & Tana, 2016). The most frequently found decrease in the physical quality of eggs is the non-optimum weight of duck eggs and thin eggshells. The size of the eggs of the Pengging ducks is smaller than those of the Magelang ducks (Suselowati *et al.*, 2019). Pavlovski *et al.* (2012) reported that around 6% – 20% of egg damage from total egg production is caused by a decrease in shell quality which causes economic losses. Rika *et al.* (2019) reported that duck eggs are prone to changes in the condition of the contents of the eggs such as a decrease in the viscosity of the white and egg yolk which has an impact on decreasing the quality of the weight of the yolk and the weight of the egg white.

The physical quality of the eggs is greatly determined by the quality of the feed given. The nutritional requirements of laying ducks are protein (17%), fat (3.5% – 6%), metabolic energy (2,500 kcal/kg), crude fiber (7%), and calcium (3% – 4%) (Fouad *et al.*, 2018). Providing additional feed ingredients in the form of feed additives can improve feed quality (Windoro *et al.*, 2020). Feed additives are given to optimise the metabolism and growth of livestock (Sunarno *et al.*, 2021). Puvača *et al.* (2020) reported that feeding additives like antibiotics did not improve egg quality. The use of antibiotics in animal feed is ineffective because it did not improve egg quality and also cause bacterial resistance, so it is necessary to look for other alternatives from raw materials that are easy to obtain, made from natural ingredients, have low prices, do not cause side effects, and are environmentally friendly, such as nanochitosan.

Nanochitosan is a polysaccharide derived from chitin, the shell of crustaceans, such as crabs and shrimp (Logesh *et al.*, 2012). Nanochitosan is a very reactive polymer due to the presence of amine groups (-NH₂) and hydroxyl groups (OH), has a positive ion charge that can increase the affinity for negative charges on organic materials, such as proteins, fats, and cholesterol (Liaqat & Eltem, 2018), Nanochitosan increases the catalytic activity of digestive enzymes by forming cross-links between the amine groups of nanochitosan and the hydroxyl groups of enzymes (Wahba, 2017). Nanochitosan has antibacterial properties which can inhibit the growth of pathogenic bacteria and encourage the growth of intestinal microflora which plays a role in improving intestinal health (Zhang *et al.*, 2020).

Sunarno *et al.* (2021) reported that giving nanochitosan with a concentration of 2.5% – 10% in feed could improve the digestive performance and productivity of Tegal ducks. Sahara *et al.* (2019) reported that providing nanochitosan in feed with a level 0.5% could increase the HU value of duck eggs by maintaining the internal quality of duck eggs, especially in egg whites, lowering egg cholesterol and increasing duck egg weight. Hamady and Farroh (2020) reported, feeding laying hens with 50 mg/kg nanochitosan produced egg with HU value 85.77.

Nanochitosan has not been widely studied as a feed additive in breeding ducks, more specifically regarding the physical quality of eggs. Therefore, this study was conducted to analyse the effect of nanochitosan as a feed additive on the physical quality of eggs as indicated by the values of HU, yolk index, egg yolk weight, egg white weight, egg weight, shell thickness, and egg shape index. This research is expected to improve the physical quality of duck eggs by adding nanochitosan as a feed additive.

MATERIALS AND METHODS

Research Plan

This study used a completely randomised design with five groups, each containing five ducks per group. Treatment with nanochitosan addition in feed was given to 20 pengging ducks. The feed groups were as follows: P0, as the control group (100% standard feed); and P1, P2, P3, and P4, representing the treatment groups with the addition of the feed additive nanochitosan to the standard feed at levels of 2.5, 5, 7.5, and 10 g/kg of feed, respectively. In this study, the independent variable is the concentration of nanochitosan, and the dependent variables are HU, IKT, and egg yolk weight, egg white weight, egg weight, shell thickness, and egg shape index.

Feed Manufacturing with Addition of Nanochitosan Feed Additives

A standard feed of 20 kg (15 kg of bran and 5 kg of concentrate) is made for five days of feeding. Every 4 kg of feed per day was given to 25 ducks. The treatment groups were given the addition of feed additive nanochitosan. Nanochitosan is a polysaccharide derived from chitin, which is found in the shells of crustaceans, such as crabs and shrimp. The nanochitosan used in this research is commercial nanochitosan. The predetermined level of nanochitosan was added to every 1 kg of standard feed weight and stirred until homogeneous. A homogeneous feed weighing 400 g was provided to each group of five ducks. Duck feed was given as much as 80 g/head with two feedings, so one duck was given 160 g feed daily. Feeding as much as 400 g was calculated based on feed conversion and the energy requirements of the ducks. According to the United States Department of Agriculture (2023),

laying ducks usually consume around 120 g of feed daily, although the amount can vary depending on production levels and the availability of feed ingredients. Duck feed was calculated as approximately 10% of the duck's weight. In this study, the ducks had an average weight of around 1.2 kg, indicating that a feed allocation of 160 g per duck/day is considered sufficient. The feed given to the ducks was a

semi-wet mash (400 g of standard dry feed mixed with 135 ml of water/400 g of feed) mixed with nanochitosan. The feed box was cleaned thoroughly every day before feeding, followed by disinfection through soaking in disinfectant solution for 30 minutes. Then, the box was dried to prevent the feed from becoming moldy. The diet formulation table is presented in Table 1 (Sunarno *et al.*, 2021).

Table 1. Nutrient composition of feed content

Feed ingredients	Nanochitosan level g/kg feed weight				
	0	2.5	5	7.5	10
Metabolic energy (Kcal)	2,630.5	2,680.9	2,790.57	2,840.8	2,880.4
Crude protein (%)	17.22	17.56	18.30	19.56	20.08
Fat (%)	6.16	5.40	5.25	4.25	4.16
Calcium (%)	1.82	2.05	2.56	2.90	3.05
Crude fiber (%)	3.07	3.25	3.57	4.09	4.21

Housing and Feeding

The ducks were placed into five plots of cages to be acclimated for one week. Each cage plot measured 100 × 150 × 70 cm³ and was equipped with a place to feed and drink. The cages were partitioned using bamboo fences, and each cage plot contains five ducks. The cages for this study used a litter system with rice husk bedding. The ducks were fed twice daily, at 8.00 a.m. and 4.00 p.m. The amount of feed for each cage with five ducks was 400 g for each feed. The feed treatments were given according to the levels: P0 (100% standard feed), and P1, P2, P3, and P4 (with the addition of nanochitosan at levels of 2.5, 5.0, 7.5, and 10.0 g/kg of feed, respectively). Water was provided ad libitum for eight weeks, starting for ducks aged 21 weeks to 28 weeks.

Variables Measurement Observation

Pengging duck eggs were collected from each cage plot every morning during the treatment period. Each duck egg was marked with a marker, namely the type of treatment and the date of collection, and collected into the egg container. Five duck eggs per treatment group were used for egg quality analysis. Measurement of research variables is carried out as follows:

Egg weight

The egg weight was obtained by weighing using a digital scale with a sensitivity of 0.1 g (Purwati *et al.*, 2015).

Egg shape index measurement

The egg shape index is measured using Eq. (1) as defined by Marcelina *et al.* (2020):

$$\text{The egg shape index} = \frac{\text{egg short axis (mm)}}{\text{egg long axis (mm)}} \times 100\% \quad \text{Eq. (1)}$$

Weight of the yolk and egg white

The weight of the yolk and egg white was obtained by breaking the egg and separating the white and yolk using an egg separator. The separated yolk and egg white were then placed in separate containers. Each container was weighed using a digital balance (Marcelina *et al.*, 2020).

Eggshell thickness

The eggshell thickness was measured using a caliper on three different sides of the eggshell: the taper, middle and blunt sides). The results were then averaged to determine the average eggshell thickness (Maskur *et al.*, 2018).

Haugh unit (HU)

The HU value is obtained using Eq. (2) as defined by Marcelina *et al.* (2020):

$$HU = 100 \log \log (H + 7.57 - 1.7W^{0.37}) \quad \text{Eq. (2)}$$

where HU = haugh unit, H = egg white height (mm), W = egg weight (g).

Egg yolk index (IKT)

The yolk index is calculated using the Eq. (3) as defined by Nuraeni *et al.* (2019):

$$IKT = \frac{\text{yolk height (mm)}}{\text{yolk diameter (mm)}} \quad \text{Eq. (3)}$$

Data Analysis

The data were analysed to determine the distribution pattern and homogeneity. The

results of the data analysis in the study showed a normal and homogeneous distribution pattern for all variables, then continued with ANOVA at a 95% confidence level. If the ANOVA results showed a significant difference, the Duncan's Multiple Range Test was conducted at a 95% confidence level to determine differences between treatment groups. This test was performed using Statistical Product of Service Solution (SPSS) version 26.0.

RESULTS AND DISCUSSION

Based on the results of the ANOVA at a 95% confidence level on the physical quality data of the eggs, it was found that the feed additive nanochitosan had a significant effect ($p < 0.05$) on the haugh value of egg units (HU), yolk index (IKT), egg yolk weight, egg white weight, egg weight, eggshell thickness, and egg shape index. The average values of these variables in the physical quality of eggs are shown in Table 2.

Table 2. Average Haugh unit (HU), yolk index (IKT), egg yolk weight, egg white weight, egg weight, shell thickness, and egg shape index of ducks after treatment with nanochitosan as a feed additive for 4 weeks

Variable	Nanochitosan treatment				
	P0 (control)	P1 2.5 g/kg	P2 5.0 g/kg	P3 7.5 g/kg	P4 10.0 g/kg
Haugh unit (HU)	72.2 ^a ±1.30	74.6 ^b ±1.14	77.2 ^c ±1.30	79.2 ^d ±1.84	81.6 ^e ±2.07
Yolk index (IKT)	0.41 ^a ±0.01	0.42 ^a ±0.02	0.44 ^b ±0.02	0.47 ^c ±0.01	0.49 ^d ±0.01
Egg yolk weight (g)	19.7 ^a ±0.51	20.3 ^{ab} ±1.15	21.2 ^b ±0.43	22.4 ^c ±0.73	24.6 ^d ±0.72
Egg white weight (g)	30.0 ^a ±1.11	31.3 ^b ±1.14	32.6 ^c ±0.53	33.9 ^d ±0.70	36.3 ^e ±0.69
Egg weight (g)	59.4 ^a ±0.79	61.1 ^b ±1.81	62.8 ^c ±0.77	64.8 ^d ±1.16	70.5 ^e ±1.10
Eggshell thickness (mm)	0.31 ^a ±0.02	0.31 ^a ±0.03	0.33 ^a ±0.03	0.39 ^b ±0.03	0.44 ^c ±0.03
Egg shape index (%)	69.2 ^a ±1.30	71.6 ^{ab} ±1.52	72.8 ^b ±2.77	76.2 ^c ±3.42	81.2 ^d ±1.92

Note: The data in the table is the average ± standard deviation. Significant differences between groups are denoted by different superscript letters within the row ($p < 0.05$). P0 - ducks fed standard feed without nanochitosan addition. P1, P2, P3, and P4 were ducks fed standard feed with nanochitosan addition of 2.5, 5.0, 7.5, and 10.0 g/kg of feed weight.

Haugh Unit (HU)

Nanochitosan had a significantly different effect on the haugh value of egg units (HU) ($p < 0.05$). Based on the data in Table 2, the value of this variable is classified as good quality. HU value is considered good quality at HU value > 72 (United States Department of Agriculture, 2000). The results of the Duncan's test showed that treatments P1, P2, P3, and P4 had higher HU values compared to P0. As the levels of nanochitosan increased, the HU values also increased. These conditions indicate that the

addition of nanochitosan as a feed additive at levels of 2.5, 5, 7.5, and 10 g/kg of feed effectively increases the HU value of eggs. Nanochitosan has the ability to increase egg protein levels due to its role as a protease activator. It contains functional groups such as the amine group (-NH₂) and hydroxyl group (-OH). These functional groups allow a crosslinking mechanism between nanochitosan and protease, involving the amine group (-NH₂) in nanochitosan with the hydroxyl group (-OH) in protease (Rosyada *et al.* 2019). This condition is characterised by the expansion of the enzyme's

surface, which increases the opportunities for interaction between the enzyme and the substrate. As a result, reduction products from proteins, such as smaller polypeptides, oligopeptides, peptides, or amino acids, are produced. The end product of protein digestion will produce amino acids, which will then be absorbed in the intestinal villi for egg formation. Lestari (2020) stated that proteases hydrolyse protein peptide bonds into small polypeptides such as amino acids. The HU value is obtained from the height and weight of the egg white, where the main component forming the egg white and egg weight is protein. Proteases are enzymes that play a role in the hydrolysis of proteins, converting them into simpler compounds like peptides and amino acids. The protein and amino acid content greatly affects the process of egg formation. The more amino acids produced, the better the quality of the egg white and egg weight which will affect the HU value.

Yolk Index (IKT)

Nanochitosan had a significantly different effect on the egg yolk index (IKT) ($p < 0.05$). Based on the data in Table 2, the value of this variable is within the normal range. Swacita and Cipta (2011) stated that fresh eggs have an IKT value ranging from 0.33 to 0.52. A good IKT score has

a value of >0.42 . The results of Duncan's test at the 95% confidence level showed that P2, P3, and P4 resulted in higher IKT values compared to P0. These conditions indicate that nanochitosan, as a feed additive at levels of 5, 7.5, and 10 g/kg of feed, effectively increases IKT values. The value of the IKT variable is obtained from the comparison between the yolk height and the yolk's diameter. The comparison of yolk height and the yolk's diameter among P0 – P4 is shown in Figure 1 and Figure 2. The purpose of determination the IKT is to assess the quality and shape of the yolk, where a higher IKT value indicates good quality while a lower value suggests the yolk is not fresh. The protein content affects the quality of the yolk in the process of forming the yolk material (vitellogenin) in the form of lipoprotein synthesis in the liver (Alfiyah *et al.*, 2015). Nanochitosan plays a role in increasing protease activity in degrading protein substrates into amino acids. Wang *et al.* (2014) stated that nanochitosan increased the catalytic activity of proteases, facilitating the degradation of proteins into amino acids for lipoprotein synthesis in the liver, which contributes to egg yolk formation. These synthesised lipoproteins accumulate in the follicles of the ovaries through the bloodstream. As the follicle matures and becomes ready for ovulation, it enters the infundibulum for egg white secretion.

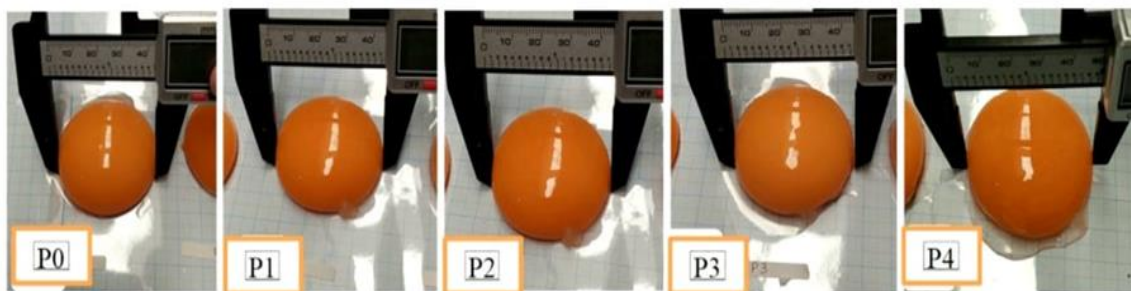


Figure 1. Diameter of egg yolk after treatment with feed additive nanochitosan for 4 weeks

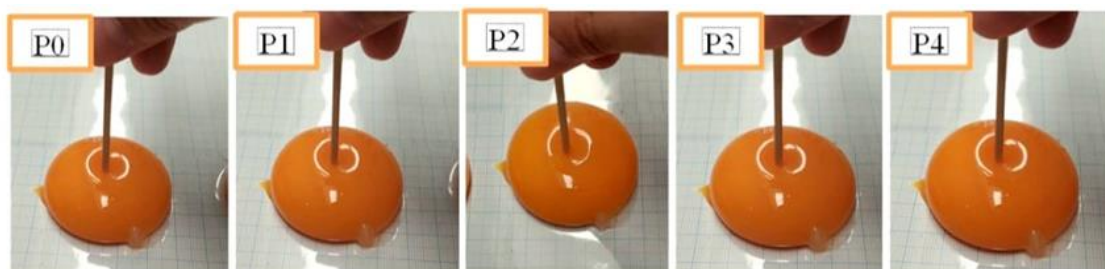


Figure 2. Height of egg yolk after treatment with feed additive nanochitosan for 4 weeks
Egg Yolk Weight

Nanochitosan had a significantly different effect on the egg yolk weight ($p < 0.05$). Based on the data in Table 2, the value of this variable is within the normal range. The percentage of egg yolk ranged from 30% – 35% of total egg weight. For duck eggs, the yolk weight usually ranges from 20 to 26 g (Ismoyowati & Purwantini, 2013). The results of the Duncan's test showed that treatments P2, P3, and P4 resulted in higher egg yolk weights compared to P0. The comparison of the egg yolk sizes among P0-P4 is shown in Figure 3. These conditions indicate that nanochitosan, as a feed additive at levels of 5, 7.5, and 10 g/kg of feed, effectively increases the egg yolk weight. The increase in egg yolk weight is thought to occur due to an increase in the protein content in the yolk. Nanochitosan

plays a role in increasing the catalytic activity of protease. Rahmawati *et al.* (2020) stated that the degradation of protein substrates into reduction products in the form of amino acids will then be absorbed by the intestinal villi for lipoprotein synthesis (egg yolk material) in the liver. Wahba (2017) stated that nanochitosan acts as a protease activator by forming cross-links between the amine groups in nanochitosan and the hydroxyl groups in proteases. This condition can increase the degradation of protein substrates in the form of amino acids needed for the formation of egg yolks. Ismoyowati and Purwantini (2013) stated that apart from fat, egg yolks also contain protein, water, and minerals which as essential components.

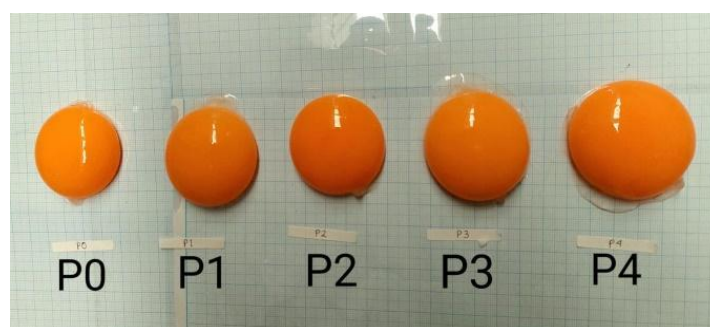


Figure 3. Comparison of the size of the egg yolks after treatment with feed additive nanochitosan for 4 weeks

Egg White Weight

Nanochitosan had a significantly different effect on egg white weight ($p < 0.05$). Based on the data in Table 2, the value of this variable is within the normal range. The percentage of egg white is about 52.6% of the egg weight. Egg white weight usually ranges from 31-38 g (Ismoyowati & Purwantini, 2013). The results of Duncan's test showed that the treatments P1, P2, P3, and P4 resulted in higher egg white weight compared to P0. These conditions indicate that nanochitosan, as a feed additive at levels of 2.5, 5, 7.5, and 10 g/kg of feed, effectively increases egg white weight. Nanochitosan treatment increased egg white weight, which was related to the increased degradation of proteins due to the increased protease catalytic activity by nanochitosan. This led to the production of more protein reduction products in the form of amino acids, which were produced and absorbed by the intestinal villi for the formation of egg white in the magnum. Lan *et al.* (2020) stated that nanochitosan increases protein digestibility by

increasing protease catalytic activity. Divya *et al.* (2017) stated that nanochitosan has antibacterial properties that can inhibit the growth of pathogenic bacteria. This condition can increase the amount of microflora in the intestine. The positive charge on nanochitosan binds to the negative charge on the cell wall of pathogenic bacteria which disrupts the permeability of the plasma membrane, then binds to DNA which leads to inhibition of protein synthesis which can inhibit the growth and reproduction of bacteria. Nugraha *et al.* (2021) stated that the beneficial microflora in the intestine improves the health and immune system of the intestine, optimising digestive activity and absorption of nutrients in the small intestine. The presence of beneficial intestinal microflora in the intestine contributes to optimising the digestion of feed and absorption of nutrients, especially the amino acids needed in the formation of egg whites. The more amino acids absorbed in the duodenum, the heavier the egg white produced.

Total Egg Weight

Nanochitosan had a significantly different effect on egg weight ($p < 0.05$). Based on the data in Table 2, the value of this variable is classified as good quality. Based on the National Standardization Agency (2008), the best quality duck eggs weigh over 60 g. The results of Duncan's test at the 95% confidence level showed that the treatments P1, P2, P3, and P4 resulted in higher total egg weight compared to P0. The comparison of egg sizes among P0 – P4 is shown in Figure 4. These conditions indicate



Figure 4. Morphology of duck egg after treatment with nanochitosan feed additive for 4 weeks

Eggshell Thickness

Nanochitosan had a significantly different effect on eggshell thickness ($p < 0.05$). The results of Duncan's test showed that P3 and P4 resulted in higher eggshell thickness than P0. The comparison of P0 – P4 eggshell thickness is shown in Figure 5. These conditions indicate that nanochitosan as a feed additive at levels of 7.5 and 10 g/kg of feed is effective in increasing eggshell thickness. The eggshell formation is strongly influenced by the availability of calcium in the feed. Calcium metabolism requires protein through the mechanism of CaBP or Calcium Binding Protein (Prabahandari *et al.*,

that nanochitosan as a feed additive at 2.5, 5, 7.5, and 10 g/kg of feed effectively increases egg weight. Egg weight is influenced by yolk weight and egg white weight. Ipek and Sozcu (2017) stated that the formation of egg yolk and egg white is strongly influenced by the effectiveness of absorption of nutrients from feed, especially protein. Sahara *et al.* (2021) stated that treatment with nanochitosan at a level of 1.5% increased protein digestibility higher than the control. Increased protein absorption in the intestine optimises the formation of egg yolks and egg whites, so egg weight also increases.

2021). Jiang *et al.* (2013) stated that the availability of high levels of calcium in the feed would improve the quality of the egg shells. Sahara *et al.* (2021) stated that nanochitosan increases protein digestibility thereby increasing the absorption of minerals and calcium for the formation of eggshells in the intestinal mucosa. Syafitri *et al.* (2015) stated that protein intake affects calcium mass in the eggshell because it plays a role in transporting calcium, namely the presence of calcium-binding protein (CaBP) or calbindin bonds, which play a role in transporting calcium into the intestinal mucosal cells in the duodenum.

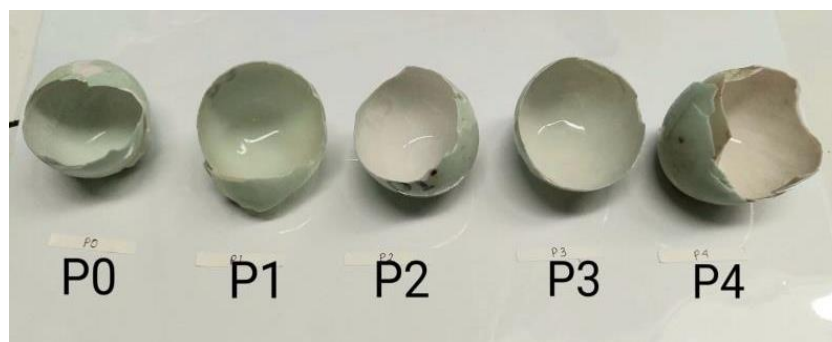


Figure 5. Eggshell after treatment with nanochitosan feed additive for 4 weeks

Egg Shape Index

Nanochitosan had a significantly different effect on the egg shape index ($p < 0.05$). Based on the data in Table 2, the value of this variable is within the normal range. Suselowati *et al.* (2019) stated that the pengging duck egg shape index was 71.84% – 86.45% and categorised as normal. Duman *et al.* (2016) stated that the ideal egg shape index value ranged from 72% – 76%. The results of Duncan's test showed that P2, P3, and P4 resulted in a higher egg shape index than P0. The comparison of P0 – P4 egg shape index is shown in Figure 6. These conditions indicate that nanochitosan as a feed additive at levels of 5, 7.5, and 10 g/kg of feed increased the value of the egg shape index. Egg size and egg weight affect the egg shape index value. The ideal egg

shape is oval (Duman *et al.*, 2016). Treatment with nanochitosan increased the value of the egg shape index, and this was due to the positive effect of nanochitosan in optimising digestive activity, especially increasing protease catalytic activity. Isik (2020) stated that nanochitosan increases the catalytic activity of enzymes by forming cross-links between the amine groups of nanochitosan and the hydroxyl groups in proteases. The value of the egg shape index can also be affected by differences in the size of the reproductive organs of ducks, such as differences in the size of the isthmus and uterus in each duck. Dirgahayu *et al.* (2016) stated that the isthmus and uterus with a wide diameter tend to produce eggs with a round shape, while the isthmus and uterus with a narrow diameter tend to produce eggs with an oval shape.



Figure 6. Egg shape index after treatment with nanochitosan feed additive for 4 weeks

Nanochitosan plays a role in immobilising proteases by forming cross-links between nanochitosan and proteases which have an important role in increasing enzyme stability and catalytic activity on protein substrates into derivative products in the form of amino acids needed for egg formation. Xin-Yan *et al.* (2012) reported that nanochitosan treatment with a level of 160 mg/kg of feed increased the activity of the trypsin enzyme in the small intestine compared to controls. Enzyme immobilisation with nanochitosan will increase the surface area and active sites of enzymes for attachment to digestive substrates so that more products are produced, as well as optimise enzyme work and allow enzymes to be reused for a longer time.

The novelty of this research is about the use of nanochitosan as a feed additive to improve the physical quality of eggs in laying ducks. This study is expected to open opportunities for further research in the future about the mechanistic effects of nanochitosan in improving egg quality and productivity of pengging ducks. The benefit of this research is

the use of nanochitosan as a feed additive to improve the physical quality of eggs especially, the effect of nanochitosan to increase the haugh unit (HU), yolk index (IKT), yolk weight, white weight, egg weight, shell egg thickness, and egg shape index in laying ducks. This research can provide a reference for farmers that nanochitosan is safe to use as a feed additive and regarding the levels of using nanochitosan as a feed additive in improving the quality of duck eggs.

CONCLUSION

The addition of nanochitosan levels improves the quality of duck eggs. The use of nanochitosan as a feed additive with a level of 2.5, 5, 7.5, and 10 g/kg feed weight can improve the quality of duck eggs as indicated by an increase in haugh unit (HU) value, egg yolk index (IKT), egg yolk weight, egg white weight, total egg weight, shell thickness, and egg shape index. Nanochitosan with levels of 7.5 and 10 g/kg feed weight produced the best effect on the quality of duck eggs.

ACKNOWLEDGEMENTS

We thank the Directorate of Research and Services Society (DRPM), Ministry of Research, Technology, and Higher Education Leading Applied Research Funding Universities in the fiscal year 2021 for the research funding.

REFERENCES

- Alfiyah, Y., Praseno, K. & Mardiyati, S.M. (2015). Indeks Kuning Telur (IKT) dan Haugh Unit (HU) telur itik lokal dari beberapa tempat budidaya itik di Jawa. *Anatomi dan Fisiologi*, 23(2): 7-15.
- National Standardization Agency (BSN). (2008). Telur Itik Konsumsi. BSN, Jakarta.
- Dirgahayu, F.I., Septinova, D. & Nova, K. (2016). Perbandingan kualitas eksternal telur ayam ras strain isa brown dan lohmann brown. *Jurnal Ilmiah Peternakan Terpadu*, 4(1): 1-5.
- Divya, K., Vijayan, S., George, T.K. & Jisha, M.S. (2017). Antimicrobial properties of chitosan nanoparticles: Mode of action and factors affecting activity. *Fibers and Polymers*, 18: 221-230.
- Duman, M., Şekeroğlu, A., Yıldırım, A., Eleroğlu, H. & Camcı, Ö. (2016). Relation between egg shape index and egg quality characteristics. *European Poultry Science*, 80: 1-9.
- Fouad, A.M., Ruan, D., Wang, S., Chen, W., Xia, W. & Zheng, C. (2018). Nutritional requirements of meat-type and egg-type ducks: what do we know? *Journal of Animal Science and Biotechnology*, 9(1): 1-11.
- Gibran, M.A., Djaelani, M.A., Kasiyati & Sunarno (2021). Bobot karkas dan morfometri serabut muskulus pektoralis itik pengging periode layer setelah pemberian tepung daun kelor dalam pakan. *Buletin Anatomi dan Fisiologi*, 6(2): 167-174.
- Hamady, G.A.A. & Farroh, K.Y. (2020). Effects of adding nano-chitosan on productive performance of laying hens. *Egyptian Journal of Nutrition and Feeds*, 23(2): 321-336.
- Ipek, A. & Sozcu, A. (2017). Comparison of hatching egg characteristics, embryo development, yolk absorption, hatch window, and hatchability of Pekin Duck eggs of different weights. *Poultry Science*, 96(10): 3593-3599.
- Isik, M. (2020). High stability of immobilized acetylcholinesterase on nanochitosan beads. *Chemistry Select*, 5(15): 4623-4627.
- Ismoyowati & Purwantini, D. (2013). Produksi dan kualitas itik lokal di daerah sentra peternakan itik. *Jurnal Pembangunan Pedesaan*, 13(1): 11-16.
- Jiang, S., Cui, L., Shi, C., Ke, X., Luo, J. & Hou, J. (2013). Effects of dietary energy and calcium levels on performance, egg shell quality and bone metabolism in hens. *The Veterinary Journal*, 198(1): 252-258.
- Kasiyati, Djaelani, M.A. & Sunarno. (2019). Effect of supplementation of *Moringa oleifera* leaf powder on reproductive performance and ovarian morphometry of pengging duck. *International Journal of Poultry Science*, 18(7): 340-348.
- Kasiyati, Djaelani, M.A. & Sunarno. (2021). Respons hematologi itik pengging yang diberi tepung daun kelor (*Moringa oleifera* Lam.) sebagai imbuhan pakan. *Jurnal Veteriner*, 22(1): 8-15.
- Lan, R., Li, Y., Chang, Q. & Zhao, Z. (2020). Dietary chitosan oligosaccharides alleviate heat stress-induced intestinal oxidative stress and inflammatory response in yellow-feather broilers. *Poultry Science*, 99(12): 6745-6752.
- Lestari, E., Sunarno, Kasiyati & Djaelani, M.A. (2020). Efek bahan aditif tepung kelor terhadap biomassa organ visceral ayam petelur jantan. *Media Bina Ilmiah*, 14(9): 3215-3229.
- Liaqat, F. & Eltem, R. (2018). Chitoooligosaccharides and their biological activities: A comprehensive review. *Carbohydrate Polymers*, 184: 243-259.
- Logesh, A., Thillaimaharani, K., Sharmila, K., Kalaiselvam, M. & Raffi, S. (2012). Production of chitosan from endolichenic fungi isolated from mangrove environment and its antagonistic activity. *Asian Pacific Journal of Tropical Biomedicine*, 2(2): 140-143.
- Marcelina, N., Djaelani, M.A., Sunarno & Kasiyati. (2020). Bobot telur, indeks bentuk telur, dan nilai kantung udara telur itik pengging setelah pemberian imbuhan tepung daun kelor (*Moringa oleifera* Lam.) dalam pakan. *Jurnal Biologi Tropika*, 3(1): 1-7.
- Maskur., Prasetyo, S., Lestari., Jan, R., Rozi, T. & Kasip, L.M. (2018). Produksi dan kualitas telur itik lokal lombok untuk bahan telur asin yang dipelihara peternak secara intensif di lombok tengah. *Jurnal Ilmu dan Teknologi Peternakan Indonesia*, 4(1): 199-206.

- Nugraha, W.T., Pradipta, M.S.I., Pramono, P.B., Soekarno, A.S. & Kusuma, B. (2021). Identifikasi morfologi mikroflora pada saluran pencernaan itik Magelang. *Jurnal Sain Peternakan Indonesia*, 16(2): 142-147.
- Nuraeni, S., Djaelani, M.A., Sunarno & Kasiyati (2019). Nilai Haugh Unit (HU), Indeks Kuning Telur (IKT) dan pH telur itik pengging setelah pemberian tepung daun kelor (*Moringa oleifera* Lam.). *Buletin Anatomi dan Fisiologi*, 4(2): 107-115.
- Pavlovski, Z., Škrbić, Z., Lukić, M., Vitorović, D., Lilić, S. & Petrićević, V. (2012). Shell quality-everlasting problem in the today poultry science. *Biotechnology in Animal Husbandry*, 28(3): 393-404.
- Prabahandari, K.A., Kasiyati, Djaelani, M.A. & Sunarno. (2021). Somatometri tulang ekstremitas ayam jantan setelah pemberian tepung daun kelor (*Moringa oleifera*) sebagai pakan tambahan. *Buletin Anatomi dan Fisiologi*, 6(2): 183-192.
- Purwati, D., Djaelani, M.A. & Yuniwati, E.Y.W. (2015). Indeks Kuning Telur (IKT), Haugh Unit (HU) dan bobot telur pada berbagai itik lokal di Jawa Tengah. *Jurnal Akademika Biologi*, 4(2): 1-9.
- Puvača, N., Lika, E., Tufarelli, V., Bursić, V., Pelić, D.L., Nikolova, N., Petrović, A., Prodanović, R., Vuković, G., Lević, J. & Glannenas, L. (2020). Influence of different tetracycline antimicrobial therapy of *Mycoplasma (Mycoplasma synoviae)* in laying hens compared to tea tree essential oil on table egg quality and antibiotic residues. *Foods*, 9(5): 1-15.
- Rahmawati, D., Djaelani, M.A., Kasiyati & Sunarno (2020). Bobot karkas dan bagian karkas ayam petelur jantan (*Gallus gallus domesticus* L.) setelah pemberian tepung daun kelor (*Moringa oleifera* Lam.) sebagai imbuhan pakan. *Jurnal Biologi Tropika*, 3(2): 65-72.
- Rika, R., Wijana, I.W. & Dewi, G.A.M.K. (2019). Kualitas eksternal dan internal telur itik yang disimpan selama 0 – 14 hari di dataran tinggi Bedugul. *Jurnal Peternakan Tropika*, 7(1): 851-863.
- Rosyada, A., Sunarharum, W.B. & Waziroh, E. (2019). Characterization of chitosan nanoparticles as an edible coating material. *IOP Conference Series: Earth and Environmental Science*, 230: 012043.
- Sahara, E., Sandi, S. & Yossi, F. (2019). The effect of chitosan and bran fermentation on the weight of abdominal fat, blood cholesterol and local duck eggs. *IOP Conference Series: Earth and Environmental Science*, 347: 012072.
- Sahara, E., Sandi, S., Yosi, F., Sari, M.L., Riswandi & Agustina, R. (2021). The potential of pure chitosan in increasing eggshell thickness silver Arabic chicken (*Silver brakel kriel*). *IOP Conference Series: Earth and Environmental Science*, 810: 012006.
- Saraswati, T.R. & Tana, S. (2016). Effect of turmeric powder supplementation to the age of sexual maturity, physical, and chemical quality of the first Japanese quail's (*Coturnix japonica*) egg. *Biosaintifika*, 8(1): 18-24.
- Sunarno, Solikhin, S. & Budiraharjo, K. (2021). Histomorphometry of the duodenum of ducks (*Anas platyrhynchos*) after administration of nanochitosan in feed. *Biosaintifika*, 13(3): 267-274.
- Suselowati, T., Kurnianto, E. & Kismiati, S. (2019). Hubungan indeks bentuk telur dan surface area telur terhadap bobot telur, bobot tetas, persentase bobot tetas, daya tetas dan mortalitas embrio pada itik pengging. *Sains Peternakan*, 17(2): 24-30.
- Swacita, I.B.N. & Cipta, I.P.S. (2011). Pengaruh sistem peternakan dan lama penyimpanan terhadap kualitas telur itik. *Buletin Veteriner Udayana*, 3(2): 91-98.
- Syafitri, Y.E., Yuniyanto, V.D. & Suthama, N. (2015). Pemberian ekstrak daun beluntas (*Pluchea indika* Less.) dan klorin terhadap massa kalsium dan massa protein daging pada ayam broiler. *Animal Agriculture Journal*, 4(1): 155-164.
- United States Department of Agriculture. (2000). Egg-Grading Manual. Department of Agriculture, Washington.
- United States Department of Agriculture. (2023). Feeding Ducks for Egg Production in Small Flocks. Department of Agriculture, Washington.
- Wahba, M.I. (2017). Chitosan-glutaraldehyde activated calcium pectinate beads as a covalent immobilization support. *Biocatalysis and Agricultural Biotechnology*, 12: 266-274.
- Wang, S., Zhang, C., Qi, B., Sui, X., Jiang, L., Li, Y., Wang, Z., Feng, H., Wang, R. & Zhang, Q. (2014). Immobilized alcalase alkaline protease on the magnetic chitosan nanoparticles used for soy

- protein isolate hydrolysis. *European Food Research and Technology*, 239(6): 1051-1059.
- Windoro, D.P.J., Kasiyati, K., Djaelani, M.A. & Sunarno, S. (2020). Pengaruh imbuhan tepung daun kelor (*Moringa oleifera* Lam.) pada pakan terhadap bobot beberapa organ dalam dan lemak abdominal itik pengging (*Anas platyrhynchos*). *Buletin Anatomi dan Fisiologi*, 5(2): 109-118.
- Wulandari, D., Sunarno & Saraswati, T.R. (2015). Perbedaan somatometri itik tegal, itik Magelang dan itik pengging. *Jurnal Akademika Biologi*, 4(3): 16-22.
- Xin-Yan, H., Wen-Li, D. & Qi-Chun, H., Zi-Rong, X. & Yi-Zheng, W. (2012). Changes in small intestinal morphology and digestive enzyme activity with oral administration of copper-loaded chitosan nanoparticles in rats. *Biological Trace Element Research*, 145(3): 355-360.
- Zhang, H., Chen, Y., Chen, Y., Li, Y., Jia, P., Ji, S., Zhou, Y. & Wang, T. (2020). Dietary pterostilbene supplementation attenuates intestinal damage and immunological stress of broiler chickens challenged with lipopolysaccharide. *Journal of Animal Science*, 98(1): 1-9.