
KAJIAN PENGGUNAAN ECO-ENZYME KULIT MANGGIS UNTUK PENGAWETAN TELUR BERBASIS TEKNOLOGI RAMAH LINGKUNGAN

STUDY OF THE USE OF MANGOSTEEN PEEL ECO-ENZYME FOR PRESERVING EGGS BASED ON ENVIRONMENTALLY FRIENDLY TECHNOLOGY

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Abstract. *Egg quality decreases during storage due to evaporation of air and gas from the egg and microbiological damage. This study aims to increase the shelf life of eggs using eco-enzymes from mangosteen peel. The benefits that can be obtained from this study are providing a solution for the community in extending the shelf life of eggs and can utilize environmentally friendly technology through the utilization of organic waste. The research procedure includes the stages of making eco-enzymes from mangosteen peel, eco-enzyme screening tests, egg sample selection, egg coating stages, and egg quality testing stages. Meanwhile, research variables include environmental temperature and humidity, difference in egg weight shrinkage, shell weight, shell thickness, air cavity, egg white index (IPT), egg yolk index (IPT), Haugh unit (HU), egg white and yolk pH values, and egg white and yolk moisture. Phytochemical testing shows that mangosteen peel eco-enzymes have phytochemical content suitable for preservation. Includes flavonoids, tannins, saponins, and alkaloids. The physicochemical characteristics of eggs after storage showed that the addition of mangosteen rind eco-enzyme had a significant effect ($P < 0.05$) on the characteristics of egg whites and yolks. Eco-enzyme has the potential to be an environmentally friendly natural preservative for eggs during storage.*

Keywords : *Eco-enzyme, Eggs, Environmentally friendly, Storage.*

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INTRODUCTIONS

Eggs are a highly popular source of protein, rich in nutrients, and easily absorbed by the body. According to Arunde *et al.* (2019), eggs are highly nutritious and have a good balance of protein and energy. However, over time, egg quality changes rapidly as

water and gas evaporate (Zuhri *et al.*, 2022). Storing eggs at room temperature accelerates egg quality degradation, including changes in pH, decreased egg yolk and white viscosity, and enlargement of the air cell (Suryono & Lukman, 2020).

During storage, water and CO₂ in eggs evaporate through the pores in the eggshell (Jazil *et al.*, 2013). The loss of CO₂ through the pores in the eggshell reduces the concentration of bicarbonate ions in the albumen and damages the buffer system (Widyastuti & Daydeva, 2018). In addition, the evaporation of water and gas from the egg will affect the nutritional content of the egg, especially protein. The longer the storage period, the more the egg protein decreases, namely 9.85% to 10.4% (Lestari *et al.*, 2018). In addition, Astuti *et al.* (2022) reported that egg weight loss is primarily caused by water evaporation, particularly in the albumen, and the evaporation of gases such as CO₂, NH₂, N₂, and some H₂S due to the decomposition of egg protein components. Eggshells have pores throughout their surface that allow water and gases to evaporate from the egg. This causes the egg's quality to rapidly deteriorate, rendering it unfit for consumption. This is reinforced by the statement (Widyastuti & Daydeva, 2018), which states that the most common egg damage is microbiological.

According to Lestari *et al.* (2018), egg quality is determined by the yolk index (YI), albumen index (EWI), air cell, egg weight, and haugh unit (HU). Poor-quality eggs will experience a decrease in the haugh unit value, a significant decrease in egg weight, and an increase in the air cell diameter. Furthermore, eggs stored for long periods are also characterized by an increase in egg pH. When the pH value increases to an alkaline level, the egg becomes a suitable substrate for microbial growth.

This study aims to increase the shelf life of eggs using eco-enzymes from mangosteen peel. The antimicro-

bial substances contained in mangosteen peel can kill (microbicidal) or inhibit (microbiostatic) the growth of contaminating microorganisms that cause spoilage of animal-derived food products. Furthermore, it can also reduce mangosteen peel waste in the environment. Therefore, egg preservation methods are needed to increase their shelf life. One method is the use of mangosteen peel eco-enzymes, which contain phytochemical compounds that function to coat the pores of the eggshell and prevent the entry of microorganisms into the egg (antibacterial) (Dungir *et al.*, 2012). In the era of global warming, this egg preservation method is essential because increasing environmental temperatures can accelerate egg deterioration during storage. In addition, eco-enzyme production is an environmentally friendly technology because it utilizes organic waste in its production.

MATERIALS AND METHODS

1. Materials

The equipment required for this study included egg racks, analytical scales, porcelain dishes, petri dishes, ovens, pH meters, glass tables, plastic, measuring cups, glass funnels, 8 gallons of 15L, micrometer screws, vernier calipers, scissors, knives, choppers, duct tape, glue, 600 mL bottles, trays, hoses, thermohygrometers are needed. The materials used are 960 laying chicken eggs, 24 kg of mangosteen peel, 8 kg of molasses, distilled water, water, rice washing water, and yeast. The laying chicken eggs used were obtained from the Sumber Unggas Jaya Farm, Bojonegoro. The weight of the eggs used is medium-sized chicken eggs (50-60 g) to achieve uniformity. Egg samples are also fresh eggs from

laying hens (less than 24 hours) and have the characteristics of normal shape and normal color.

2. Methods

2.1 Making Eco-enzyme from Mangosteen Peel

The making of eco-enzyme is in accordance with the modified research of (Fadlilla *et al.*, 2023). The washed and sliced mangosteen entire peel is put into a gallon along with water and brown sugar using a ratio of 1 sugar: 3 organic waste: 10 water. The making of eco-enzyme uses 4 different ingredients: E0 (water), E1 (rice washing water), E2 (water with yeast), E3 (rice washing water with yeast), then stored for 40 days. Eco-enzyme is made in 2 gallons for each treatment, so the total amount is 8 gallons. Next, eco-enzyme tests are carried out which include flavonoid, saponin, tannin, alkaloid, steroid, antibacterial, TDS, and pH tests. The best results will be used for egg dipping to coat eggshell using eco-enzyme from mangosteen peel.

2.2 Eco-enzyme Screening Test

2.2.1 pH and TDS measurements

The pH and TDS measurements were performed using an integrated digital pH meter. This allows a single instrument to measure both pH and TDS.

2.2.2 Antibacterial Activity Test

Antibacterial testing uses zones of inhibition measurements. Isolated bacteria from spoiled eggs are inoculated onto agar media. Drops of eco-enzyme are then added to form a zone of inhibition (a clear, non-growth zone) around the drop. This method applies the principle of diffusion, meaning the highest concentration of eco-enzyme is located around the drop and decreases with

distance from the drop. The wider the zone of inhibition around the drop, the stronger it is against bacteria. The diameter of the zone of inhibition is measured using a vernier caliper.

2.2.3 Flavonoid Test

The addition of Mg and HCl to the mangosteen peel eco-enzyme resulted in the formation of a red color, indicating the presence of flavonoids. A positive result is indicated by a color change from red to yellow to orange (Ramadani *et al.*, 2022).

2.2.4 Tannin Drop Test

Tannin testing is performed using a color reaction test with the addition of 1% FeCl₃. A blue-black or blackish-green color indicates the presence of tannin (Ramadani *et al.*, 2022).

2.2.5 Tannin Test Using Gelatin

The gelatin tannin test uses 3 drops of 10% gelatin. A positive test result indicates the presence of a white precipitate.

2.2.6 Alkanoid Test

Alkaloid testing can be performed using three reagents: Mayer's, Dragendorff's, and Wagner's. A positive result for alkaloids in Mayer's reagent is indicated by the presence of a white precipitate. A positive result for Dragendorff's reagent is indicated by the formation of an orange precipitate in the solution (Maharani *et al.*, 2021). Meanwhile, alkaloids in Wagner's reagent are indicated by the presence of a brown precipitate in the tube (Wibowo *et al.*, 2024).

2.2.7 Steroid Test

There are two types of steroid tests used: alcohol and non-alcohol. Place 1 mL of eco-enzyme in a cup, add 1 mL of alcohol, and evaporate the mixture until the liquid evaporates and dries. Add 5 drops of H₂SO₄ and 3 drops of acetic acid anhydrous. The formation of

a green ring indicates the presence of steroid compounds. The non-alcoholic test is performed by adding 1 mL of eco-enzyme to 3 drops of acetic acid and 3 drops of sulfuric acid (a positive result is indicated by the presence of a blue-green ring).

2.2.8 Saponin test

The addition of 1 drop of 2N HCl produces foam 1-10 cm high and persists for at least 10 minutes, indicating a positive saponin content (Maharani *et al.*, 2021).

2.3 The Administration of Eco-enzyme to Eggs

Eggs are dipped in eco-enzyme to close pores, prevent water and gas evaporation, and prevent bacterial entry (Dharmawibawa, 2022). The egg dipping method is carried out according to a modified study (Tindjabate *et al.*, 2014). Eggs are dipped in an eco-enzyme solution mixed with distilled water. The dipping is carried out until all surfaces are covered with the solution and left for 1 minute, then drained. Eggs will be stored at room temperature for 30 days with storage stages of 0, 10, 20, and 30 days. The treatment has 4 replications, each replication containing 10 eggs. The treatments are as follows:

- P0 : Control using aquadest only
- P1 : 25% eco-enzyme diluted in aquadest v/v

- P2 : 50% eco-enzyme diluted in aquadest v/v
- P3 : 75% eco-enzyme diluted in aquadest v/v
- P4 : 100% eco-enzyme diluted in aquadest v/v

2.4 Eggs Index Measurements

Egg index measurements are performed to determine uniformity. The following formula is used :

$$EI = \frac{(W1 + W2) : 2}{L \text{ (mm)}} \times 100\%$$

Note :

EI: egg index;

W1: transverse width of egg;

W2: longitudinal width of egg,

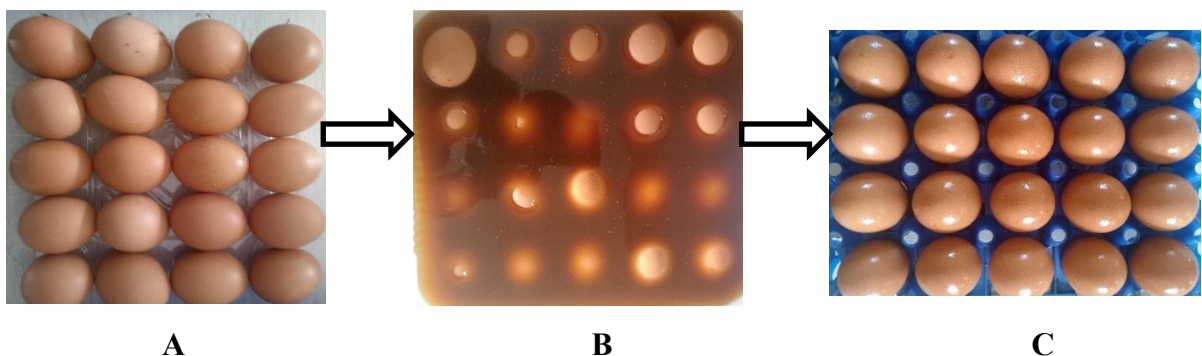
L: length of egg

2.5 Eggshell Thickness

Eggshell thickness is measured after the egg is cracked. The measured eggshells are the blunt shell and the pointed shell, which will then be averaged. Before being measured, the egg shell is first separated from the shell membrane.

2.6 Eggshell Weight

Eggshell weight is also used to measure uniformity. Each shell whose thickness has been measured will be weighed to calculate its shell weight.



Note: A: eggs before dipping; B: eggs are dipped in eco-enzyme solution; C: eggs after dipping

2.7 Egg Weight Loss

Weight was calculated before and after storage. The initial weight was measured after the eggs were removed from the cage, and the final weight was measured after dipping and storage. Weight loss was calculated to determine the percentage loss in weight of the eggs treated with eco-enzyme. The formula for calculating egg weight loss is as follows.

$$EWLD = \frac{(w1 - w2)}{w1} \times 100\%$$

Note:

- EWLD : Egg Weight Loss Difference
- w1 : initial weight (before storage)
- w2 : final weight (after storage)

2.8 Air Cell Measurements

The air cell was measured after the egg was cracked. Before measuring the eggshell thickness, the air cell was first measured. The measurement was performed using a vernier caliper with units of cm and two decimal places.

2.9 Albumen Index (AI) Measurements

Measurements are made on the thick area of the albumen. The formula for measuring the albumen index is as follows:

$$AI = \frac{H}{(W1 + W2) : 2}$$

Note:

- AI : Albumen index
- H : Thick albumen height
- W1 : Width of albumen
- W2 : Width of albumen from another part.

2.10 Yolk Index (AI) Measurements

The yolk must not be broken to allow for height and diameter measurements. The formula for measuring the yolk index is as follows:

$$\text{Yolk Index} = \frac{h}{(W1 + W2) : 2}$$

Note:

- YI : Yolk index
- H : Yolk height
- W1 : Width of yolk
- W2 : Width of yolk from another part

2.11 Haugh Unit (HU)

The HU calculation begins with measuring the egg weight and then the height of the albumen. The egg weight is measured using an analytical balance. The albumen height is measured using a vernier caliper. The formula for calculating HU is as follows:

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

- HU : Haugh unit;
- H : Thick albumen height;
- W : Egg weight.

2.12 Moisture Content of Albumen and Yolk

Weight 2 grams of separated albumen and yolk. The porcelain cup is preheated in an oven at 105°C. After the porcelain cup is placed in the oven, it is placed in a desiccator until its weight is constant. Finally, a 2 g sample is placed in the empty, pre-weighed porcelain cup and oven-dried until the weight is stable, then,

$$\text{Moisture} = \frac{w1 - w2}{w1} \times 100\%$$

Note:

- w1 : Initial weight;
- w2 : Final weight

2.13 The pH Value of Albumen and Yolk

The pH measurement is performed after the albumen and yolk are separated. The pH measurement uses a calibrated digital pH meter. The pH values of the albumen and yolk are measured separately.

2.14 Data Analysis

The research data were analyzed using a completely randomized design (CRD) with a factorial pattern. Treatments that showed a significant effect ($P < 0.05$) were then followed by a Duncan Multiple Range Test (DMRT) to determine the range of differences in the results (Duncan, 1955). The following data will be presented in the form of notations to show the differences in the interaction effect of the two factors, namely eco-enzyme concentration and storage time.

RESULTS AND DISCUSSION

1. Organoleptic, pH, and TDS Tests of Eco-enzyme

The results of the organoleptic, pH, and TDS observations are shown in Table 1. The TDS (total dissolved solids) value indicates the amounts of dissolved substances in the solution being measured. Measurements were conducted on four eco-enzyme treatments.

The color and aroma of eco-enzyme before and after fermentation did not differ significantly. This is because the eco-enzyme did not experience any leaks or other problems during storage.

Therefore, the color and aroma of the eco-enzyme did not experience significant changes. The color produced by the eco-enzyme is related to the fermentation process. The results of this study indicate that the resulting eco-enzyme product has a light to dark brown color. An eco-enzyme can be considered good if the resulting eco-enzyme produces a brown solution and a distinctive, fresh, sour aroma. The color of the eco-enzyme product also depends on the combination of fruit peel waste used. According to Surtina *et al.* (2020), eco-enzyme is a complex solution that produces a dark brown color and has a strong sweet and sour aroma. These results also align with the findings of Rusdianasari *et al.* (2021),

who showed that the resulting eco-enzyme is dark brown in color and has a fresh, sour aroma. pH measurements were conducted to determine changes in eco-enzymes. Measurements showed that the pH decreased after fermentation. Generally, the longer the fermentation time, the lower the pH value (Benny *et al.*, 2023). The low pH in eco-enzymes is caused by the high organic acid content.

Table 1. Organoleptic, pH, and TDS Tests

Sample	Color		Aroma		pH		TDS	
	Initial	End	Initial	End	Initial	End	Initial	End
E0	3	3	+	+	4.55	4.36	966	5385
E1	2	1	+++	+++	4.57	4.37	1109	5605
E2	3	2	+	++	4.45	4.25	1114	5505
E3	2	2	+++	+++	4.46	4.20	1146	5810

Description: E0: water; E1: water & yeast; E2: washing water; E3: rice washing water & yeast

Color: (1) slightly brown; (1) brown; (3) dark brown

Aroma: (+) slightly sour; (++) sour; (+++) very sour

Eco-enzymes contain organic acids in the form of acetic acid and lactic acid. This is in line with the opinion of Rasit *et al.* (2019) that low pH is determined by the presence of organic acids. Anaerobic metabolism (without oxygen) or fermentation is an effort by bacteria to obtain carbohydrate energy under anaerobic conditions and produce alcohol or acetic acid (depending on the type of microorganism). According to Larasati *et al.* (2020), the metabolic process produces acetic acid which is found in fruit and vegetable waste. Fungi and several types of bacteria produce alcohol and acetic acid during the fermentation process.

Total dissolved solids (TDS) include the total amount of minerals, salts, and organic matter dissolved in a solution. This depends on the formulation used. Initial eco-enzyme

research showed a TDS between 900 and 1200 ppm. After 40 days of storage, the eco-enzyme increased, with results shown in Table 1. Eco-enzymes made using organic materials such as fruit waste or organic solid waste and molasses as substrates in the fermentation process contribute to a high TDS factor (Rasit *et al.*, 2019).

2. Eco-enzyme Antibacterial Test

The antibacterial test aims to determine the effectiveness of eco-enzymes in inhibiting the growth of test bacteria. Inhibitory effectiveness increases with the size of the inhibition zone (clear zone). This test is to indicate that eco-enzyme could inhibit bacteria on the screening method. The best inhibition shows that eco-enzyme have potential for egg preservation.

Table 2. Eco-enzyme inhibition zone values against bacteria isolated from spoilage eggs

Sample	Test Parameter	Result (mm)	Method
E0	Inhibition Zone	8.75	Agar disk diffusion
E1	Inhibition Zone	9.75	Agar disk diffusion
E2	Inhibition Zone	10.00	Agar disk diffusion
E3	Inhibition Zone	10.76	Agar disk diffusion

Table 3. Results of qualitative phytochemical test

Phytochemical	Testing		Eco-enzyme			
	Reagents	Detection	E0	E1	E2	E3
<i>Alkaloids</i>	<i>Dragendorf</i>	Red-Yellow Sediment	-	-	+	+
	<i>Mayer</i>	White Sediment (Cream)	+	+	+	+
	<i>Wagner</i>	Purple-Yellow	+	+	+	+
<i>Tannin</i>	<i>FeCl₃</i>	Blackish-Blue	+	+	+	+
	Gelatin	White Sediment	+	+	+	+
<i>Steroids</i>	Alkohol	Green Ring	-	-	-	-
	<i>CuSO₄ & H₂SO₄</i>	Blue or Green	-	-	-	-
<i>Flavonoids</i>	<i>Mg & HCL</i>	Orange	+	+	+	+
<i>Saponin</i>	Hot Water	Stable Foam Forms	+	+	+	+

Description: (+) positive test result; (-) negative test result.

Chemical compounds such as tannins in eco-enzymes can form phenol-protein complexes with weak bonds, which then damage the cytoplasmic membrane and cause cell body leakage (Ramadani *et al.*, 2022). Nurliana & Musta (2019) stated that the higher the concentration of bioactive compounds used, the greater the ability to inhibit bacteria. The chemical inhibitory mechanism can form interactions that produce protein-phenolic complexes. The strength of inhibition depends on the concentration of the phenolic content used. The test results showed that the average inhibition zone of eco-enzymes against bacteria was the widest, namely in E3. According to Siswara *et al.* (2025), the inhibitory power of bacterial growth can be seen from the formation of a clear zone by an antibacterial compound against bacteria grown in the media. The inhibitory power in this study included moderate growth (5-10 mm). In this case, the acetic acid content in eco-enzyme plays a role in inhibiting the growth of *Streptococcus spp* bacteria (Welfalini *et al.*, 2022).

3. Eco-enzyme Phytochemical Testing

Phytochemical screening is the initial stage in identifying the chemical compounds contained in eco-enzymes. The following are the results of phytochemical screening on mangosteen peel eco-enzymes.

In general, test results show that eco-enzymes contain phytochemical compounds including alkaloids, saponins, tannins, and flavonoids. However, eco-enzymes do not contain steroids. Tannins function as antimicrobials, antioxidants, and can cover microscopic cracks, making them more resistant to physical damage during

storage. Tannins, as polyphenolic compounds, can bind to egg membrane proteins. The combination of tannins and saponins can increase egg stability and reduce contamination from bacteria and fungi. Tannins can prevent bacteria by inhibiting bacterial DNA, which prevents cell formation in bacteria (Sunani & Hendriani, 2023). Therefore, eco-enzymes can later inhibit bacterial growth, which can affect egg quality. Based on the screening results of eco-enzyme, the eco-enzyme chosen for egg preservation in this study is E3.

4. Temperature and Humidity Conditions for Egg Storage Environment

The temperature and humidity of the storage room can be seen in Table 4. Recording the temperature and humidity is necessary to understand the dynamics of changes that occur during the storage process

Eggs are generally stored at room temperature. Storing eggs at room temperature accelerates the evaporation of gas and water in the eggs, thus facilitating deterioration of egg quality. The principle of egg storage is to minimize the evaporation process that occurs in eggs. Therefore, storage at the correct temperature is necessary to slow the evaporation process (Bilyaro *et al.*, 2021).

Conditions observed during storage included temperature and humidity in the storage room. The average morning temperature was 29.81°C with a humidity of 54%. During the day, the average temperature was 31.15°C with an average humidity of 39%. Meanwhile, in the afternoon, the average temperature was 31.99°C with an average humidity of 36%.

Table 4. Temperature and humidity conditions during egg storage

Day	Temperature (°C)			Average	Humidity			Average
	1	2	3		1	2	3	
1	30.2	32.2	31.5	31.51	64%	65%	68%	66%
2	30.3	31.6	32.8	31.57	69%	64%	57%	63%
3	30.2	31.7	32.9	31.60	68%	64%	57%	63%
4	30.2	31.5	32.3	31.33	64%	63%	66%	64%
5	30	31.1	32.8	31.30	69%	64%	60%	64%
6	31.8	35.2	32.4	33.13	68%	66%	58%	64%
7	29.8	31.7	30.8	30.77	65%	67%	62%	65%
8	30.3	31.6	32.1	31.33	70%	52%	51%	58%
9	30.2	31.5	32	31.23	64%	54%	59%	59%
10	29.7	31.5	31.1	30.77	70%	67%	61%	66%
11	29.7	31.2	31.1	30.67	67%	64%	65%	65%
12	30.3	30.9	34.1	31.77	73%	69%	65%	69%
13	28.6	31.6	32.2	30.80	80%	69%	64%	71%
14	29.2	30.4	31	30.20	67%	58%	61%	62%
15	29.6	30.4	32.1	30.70	63%	51%	58%	57%
16	29.2	31.1	32.5	30.93	67%	50%	51%	56%
17	29.8	30.2	32.4	30.80	63%	51%	53%	56%
18	29.5	30.7	33.1	31.10	64%	50%	51%	55%
19	29.2	30.1	31.7	30.33	68%	59%	51%	59%
20	28.9	29.9	32	30.27	61%	48%	54%	54%
21	29.2	30.3	31	30.17	70%	62%	65%	66%
22	29.8	30.9	30.8	30.50	68%	65%	69%	67%
23	29.9	30.4	30.4	30.23	70%	61%	64%	65%
24	29.4	30.3	31	30.23	65%	56%	58%	60%
25	29.4	30.5	31.8	30.57	63%	67%	66%	65%
26	29.5	31.1	32.4	31.00	67%	72%	61%	67%
27	30.5	31.6	32.1	31.40	64%	58%	62%	61%
28	30.2	32.7	32.9	31.93	64%	52%	55%	57%
29	29.9	30.4	32.2	30.83	63%	51%	59%	58%
30	29.9	30	31.4	30.43	68%	61%	63%	64%
Average	29.81	31.15	31.99		67%	60%	67%	

Description: Temperature and humidity measurements are carried out 3 times a day (morning, afternoon, evening).

According to BSN (2008), egg shelf life is highly dependent on temperature and humidity during storage. Eggs have a shelf life of 14 days or 2 weeks when stored at room temperature and 80-90% humidity. These observations yielded varying results. This is because the amount of water vapor in the air changes daily (Sari, 2021). Due to tem-

perature differences, various biochemical processes and evaporation occur, affecting the rate of egg quality decline during storage. Meanwhile, eggs stored at low temperatures, namely 4-7°C and 60-70% humidity, can be stored for 30 days or one month (BSN, 2008). This shows that temperature and humidity significantly influence egg shelf life

5. Characteristics of the Egg Uniformity Profile in Research

The eggshell is the outermost layer, acting as a barrier against external pressure and the invasion of microorganisms. Eggshell weight and thickness can affect the shelf life of the egg. Eggshell weight and thickness are factors determined by the quality of the chicken feed. Chickens naturally regulate their eggshell formation depending on the calcium and phosphorus they consume. Furthermore, genetics and husbandry also influence eggshell

properties (Setiawati *et al.*, 2016). This study used the eggshell as the egg profile used for uniformity parameters. Egg uniformity can be seen from the eggshell, which showed no significant differences in eggshell values.

The results of the egg index, eggshell weight, and thickness measurements showed no significant difference or similar values ($P>0.05$). This indicates that the eggs used in the study had a uniform egg profile. This was used to ensure that the results were influenced solely by the treatment.

Table 5. Egg profile in the study

Eco-enzyme	Day				Average
	D0	D10	D20	D30	
	Egg Index				
P0	0.79±0.02	0.81±0.03	0,79±0,03	0,79±0,01	0,79±0,02
P1	0.80±0.01	0.81±0.03	0,79±0,01	0,78±0,02	0,79±0,01
P2	0.78±0.01	0.79±0.01	0,79±0,01	0,77±0,01	0,78±0,01
P3	0.79±0.01	0.80±0.01	0,79±0,01	0,78±0,01	0,78±0,01
P4	0.80±0.01	0.80±0.01	0,78±0,01	0,79±0,02	0,79±0,01
Average	0.79±0.01	0.79±0.01	0,78±0,01	0,78±0,01	
	Eggshell Thickness (mm)				
P0	0.46±0.08	0.37±0.02	0,45±0,05	0.43±0.06	0.42±0.05
P1	0.41±0.02	0.38±0.02	0.41±0.02	0.39±0.01	0.39±0.01
P2	0.45±0.02	0.42±0.00	0.42±0.01	0.37±0.04	0.41±0.01
P3	0.42±0.02	0.39±0.02	0.43±0.03	0.40±0.03	0.41±0.02
P4	0.40±0.02	0.36±0.01	0.41±0.02	0.39±0.01	0.39±0.01
Average	0.42 ±0.03	0.38±0.01	0.42±0.02	0.39±0.03	
	Eggshell Weight (g)				
P0	6.48±0.16	6.60±0.09	6.34±0.11	6.31±0.16	6.35±0.13
P1	6.46±0.20	6.45±0.16	6.37±0.11	6.34±0.25	6.40±0.18
P2	6.45±0.11	6.42±0.11	6.63±0.05	6.24±0.13	6.43±0.10
P3	6.46±0.20	6.46±0.04	6.43±0.11	6.48±0.09	6.43±0.11
P4	6.36±0.23	6.31±0.15	6.40±0.26	6.38±0.12	6.45±0.19
Average	6.35±0.18	6.43±0.11	6.43±0.12	6.44±0.15	

Note: Different superscripts in the same row and column indicate no significant difference ($P>0.05$).

Table 6. Results of Measurement of Physicochemical Properties and Egg White Characteristics

Eco-enzyme	Day			
	D0	D10	D20	D30
Moisture (%)				
P0	86.48 ± 1.57 ^{abcd}	86.39 ± 0.31 ^{abcd}	85.65 ± 0.51 ^{cd}	83.69 ± 0.79 ^{abcd}
P1	86.89 ± 0.19 ^{abcd}	86.80 ± 0.83 ^{abcd}	86.95 ± 0.35 ^{abcd}	84.21 ± 1.48 ^{abcd}
P2	87.71 ± 0.54 ^{ab}	86.05 ± 0.45 ^{bcd}	86.28 ± 0.46 ^{abcd}	85.06 ± 0.67 ^d
P3	87.67 ± 0.41 ^{ab}	87.09 ± 0.13 ^{abc}	86.09 ± 0.39 ^{bcd}	85.06 ± 0.97 ^d
P4	88.17 ± 0.85 ^a	87.56 ± 0.44 ^{ab}	87.11 ± 1.71 ^{abc}	84.32 ± 3.51 ^e
pH				
P0	9.54 ± 0.10 ^{ef}	10.34 ± 0.04 ^{abc}	10.40 ± 0.03 ^{abc}	10.37 ± 0.06 ^{abc}
P1	9.20 ± 0.08 ^f	10.28 ± 0.10 ^{abc}	10.48 ± 0.09 ^{ab}	10.46 ± 0.05 ^{ab}
P2	9.21 ± 0.04 ^f	10.73 ± 1.11 ^a	10.55 ± 0.01 ^{ab}	10.38 ± 0.09 ^{abc}
P3	9.18 ± 0.08 ^f	10.10 ± 0.07 ^{bcd}	10.36 ± 0.11 ^{abc}	9.28 ± 0.87 ^f
P4	9.23 ± 0.10 ^f	9.98 ± 0.05 ^{cd}	10.31 ± 0.05 ^{abc}	9.80 ± 0.17 ^{de}
Albumen Index (AI)				
P0	0.12 ± 0.01 ^{bc}	0.05 ± 0.01 ^e	0.02 ± 0.01 ^{ij}	0.02 ± 0.01 ^j
P1	0.12 ± 0.01 ^{bc}	0.05 ± 0.00 ^{efg}	0.02 ± 0.00 ^{ij}	0.02 ± 0.00 ^{ij}
P2	0.11 ± 0.01 ^c	0.05 ± 0.01 ^{efg}	0.02 ± 0.00 ^j	0.03 ± 0.01 ^{hij}
P3	0.12 ± 0.01 ^{ab}	0.05 ± 0.01 ^{ef}	0.03 ± 0.01 ^{hij}	0.03 ± 0.01 ^{ghi}
P4	0.13 ± 0.01 ^a	0.07 ± 0.01 ^d	0.04 ± 0.01 ^{fgh}	0.04 ± 0.01 ^{efgh}
Haugh Unit (HU)				
P0	92.99 ± 1.95 ^a	70.55 ± 4.78 ^{bc}	41.84 ± 11.73 ^{hi}	40.06 ± 10.26 ⁱ
P1	91.46 ± 2.38 ^a	63.32 ± 3.81 ^{cde}	48.43 ± 1.79 ^{ghi}	47.24 ± 3.61 ^{ghi}
P2	89.36 ± 3.33 ^a	65.42 ± 3.62 ^{cd}	50.00 ± 6.59 ^{fgh}	39.65 ± 7.37 ⁱ
P3	92.67 ± 1.32 ^a	66.10 ± 4.99 ^{bcd}	60.14 ± 7.21 ^{de}	54.50 ± 9.43 ^{efg}
P4	95.31 ± 3.69 ^a	74.99 ± 8.32 ^b	59.19 ± 7.41 ^{def}	55.12 ± 3.55 ^{efg}

Note: Different superscripts in the same row and column indicate a significant difference ($P < 0.05$).

6. Egg White Characteristics After Storage

The analysis of moisture data showed a significant difference ($P < 0.05$). The eco-enzyme treatment showed a higher decrease in moisture during storage. This was caused by two factors: internal shrinkage, or from the yolk, and external shrinkage. However, the decrease in moisture in egg whites occurred because the covalent bonds in the egg white had broken down, making the egg white thinner and containing more water. Meanwhile,

internal shrinkage, namely from the yolk, where the vitelline membrane of the egg had been damaged, resulted in the egg white being unable to maintain its quality and having a higher moisture content. The increase in pH affected the buffer balance, causing ovomucin to be unable to maintain the viscosity of the egg white, resulting in the egg white becoming thinner and its moisture increasing (Mangalisu *et al.*, 2021).

Egg white pH analysis showed significantly different results ($P < 0.05$). The use of 75% eco-enzyme reduced the

alkaline properties of egg whites on the 30th day, but the use of 100% eco-enzyme was lower than the 75% treatment in maintaining pH values. The increase in egg white pH is caused by a decrease in bicarbonate ions in the egg white due to the loss of CO₂ (Mangalisu *et al.*, 2021). The use of mangosteen peel eco-enzyme provides protection through tannins as a coating on the egg shell, thus preventing protein degradation caused by reduced CO₂ in the egg white. Ovomucin is not damaged, so the viscosity of the egg white is maintained.

Characteristic testing, especially on the AI, showed significantly different results ($P < 0.05$). The preservation results can be seen in P4 which was able to maintain a high AI value, this indicates that the mangosteen peel eco-enzyme has enzymes that are able to maintain albumin to stay fresh and good. AI is influenced by the viscosity of the egg white, the thicker the egg white, the better the AI value. The decrease in AI is due to the breakdown of carbonic acid into CO₂, so that the pH decreases and reduces the buffer balance (Azizah *et al.*, 2017). Ovomucin will lose its structure when the pH increases. So that the thick egg white becomes runny due to the destruction of ovomucin. If the pH and moisture indicate that the treatment is good, the AI will also follow.

The use of mangosteen peel eco-enzyme was able to maintain the HU value. This result was demonstrated by the Haugh unit analysis which produced significantly different data ($P < 0.05$). The use of eco-enzyme was better than the control treatment which showed a value of 41.84 ± 11.73 , while the 75% treatment showed a result of 60.14 ± 7.21 . The HU value follows the

AI value because in measuring HU, the height of the egg white is required, which requires the viscosity of the egg white. The HU is a value that reflects the condition of the egg albumen which is useful for determining egg quality (Azizah *et al.*, 2017). The shrinkage is caused by evaporation from within the egg and the thinness of the egg white. So if the air cavity has a high value, the HU can be low, and the thinner the egg white, the lower the HU will be.

7. Yolk Characteristics, Egg Weight Loss, and Air Cell

Research by Akyurek & Okur (2009) showed that storage time can cause mixing of egg white and yolk. Sudaryani (2003) also stated that egg storage time will experience changes in quality. This is because the vitelline membrane in the egg yolk and some of its proteins are damaged, resulting in continuous water flow from the egg white to the yolk. Analysis of the yolk index data showed a significant interaction with the results. The yolk index is the ratio of the height of the yolk to the diameter of the yolk. Normal yolk index values are between 0.33 and 0.50 (Do Espírito Santo *et al.*, 2017). Testing the yolk index on D0 showed no significant difference because there was no storage time on D0. Meanwhile, it decreased on D10-D30. This is due to water migration in the yolk. The longer the egg is stored, the lower the yolk index value.

The air cell depth < 5 mm is included in quality I, the air cavity depth of 5-9 mm is included in quality II, and the air cavity depth > 9 mm is included in quality III. Based on the explanation, the storage D0 is included in quality I, which ranges from 3.46 ± 0.45 to 3.74 ± 0.83 . From D10 to D30, it increases. The longer the storage period, the greater

the depth of the air cavity. According to research by Jazil *et al.* (2013), the air cavity is formed shortly after the egg is produced as a result of the difference in room temperature which is lower than the mother's body temperature. When the storage process is carried out, it can reduce the loss of CO₂ and water and slow down the structural changes of the albumen, the increase in osmotic pressure between the albumen and the yolk will slow down, resulting in better yolk quality during storage (Xu *et al.*, 2018).

Based on the measurement results, storage can cause egg weight loss. The decrease in egg weight is influenced by the release of CO₂ and water vapor in the albumen due to the increasingly open pores. This evaporation and gas release occurs continuously during storage, resulting in a decrease in egg weight. The average weekly egg weight loss is 2.60 g (Jazil *et al.*, 2013). According to FAO (2003), a 2-3% decrease in egg weight is still acceptable to consumers. It can be seen that in this study, the best egg storage was P4 on the 10th day, with a shrinkage of 2.06 ± 0.14. This indicates that the use of eco-enzymes has an effect on egg weight loss. The use of eco-enzymes functions to close the pores to prevent evaporation from the egg. In addition, it prevents microorganisms from entering and multiplying inside the egg, which results in a decrease in egg weight. A decrease in egg weight can cause losses in sales and the longer the eggs are

stored, the more biochemical damage the egg structure can cause (Siswara *et al.*, 2023).

The results of the egg yolk moisture analysis showed a significant interaction. The use of eco-enzymes in egg preservation had a significant effect. This was evident in each treatment, which experienced a systematic increase in moisture with storage time. This impacts the process of water evaporation from the egg to the outside of the eggshell through the pores (Suresh *et al.*, 2015).

The analysis of egg yolk pH data showed a significant interaction effect. Therefore, from D0 to D30, there was a difference in pH, namely an increase. The smallest increase in pH value after 30 days of storage occurred at P4, proving that the use of eco-enzymes can inhibit the increase in pH. Ningtiyas *et al.* (2023) stated that the pH of good quality egg yolk is 6.0. The longer the egg is stored, the more the pH increases. Factors influencing the increase in pH are due to evaporation and the loss of most of the carbon dioxide. The balance of protein, bicarbonate ions, and carbon dioxide are influential indicators of the increase in pH. As a result of the diffusion process, water moves towards the yolk, causing permeation and stretching the vitelline membrane. When permeation occurs, it can accelerate the dilution of the yolk and increase the pH of the yolk (Mangalisu *et al.*, 2021).

Table 7. Measurement results of egg weight loss, air cavity, and egg yolk characteristics

Eco-enzyme	Day			
	D0	D10	D20	D30
	Yolk Index (YI)			
P0	0.45±0.01 ^a	0.22±0.04 ^{cd}	0.17±0.01 ^{def}	0.11±0.02 ^g
P1	0.43±0.01 ^a	0.27±0.02 ^b	0.14±0.04 ^{fg}	0.09±0.01 ^g
P2	0.46±0.01 ^a	0.26±0.03 ^{bc}	0.13±0.01 ^{fg}	0.13±0.02 ^{fg}
P3	0.43±0.03 ^a	0.30±0.01 ^b	0.22±0.04 ^{cd}	0.13±0.04 ^{fg}
P4	0.45±0.01 ^a	0.31±0.02 ^b	0.20±0.03 ^{de}	0.16±0.04 ^e
	Air Cell (cm)			
P0	3.46±0.45 ^e	6.19±0.52 ^d	8.66±0.63 ^c	10.75±1.18 ^a
P1	3.38±1.01 ^e	5.98±0.21 ^d	9.75±1.17 ^b	11.54±0.81 ^a
P2	3.65±0.49 ^e	6.19±0.79 ^d	10.72±0.82 ^a	10.80±0.76 ^a
P3	4.05±0.63 ^e	6.51±0.33 ^d	9.36±1.00 ^{bc}	10.92±1.15 ^a
P4	3.74±0.83 ^e	6.05±0.44 ^d	10.67±0.93 ^a	11.04±0.32 ^a
	Egg Weight Loss (g)			
P0	0.00±0.00 ^f	2.46±0.21 ^e	4.43±0.20 ^d	7.36±0.45 ^a
P1	0.00±0.00 ^f	2.19±0.09 ^e	4.53±0.30 ^d	7.41±0.45 ^a
P2	0.00±0.00 ^f	2.39±0.24 ^e	5.06±0.27 ^c	7.54±0.94 ^a
P3	0.00±0.00 ^f	2.23±0.08 ^e	4.34±0.18 ^d	6.41±0.75 ^b
P4	0.00±0.00 ^f	2.06±0.14 ^e	4.38±0.16 ^d	6.42±0.26 ^b
	Moisture (%)			
P0	46.62±0.63 ^g	51.53±0.70 ^f	54.60±2.88 ^{cde}	54.48±0.56 ^{cd} e
P1	43.88±0.91 ^h	54.73±2.07 ^{cde}	54.96±1.44 ^{cd}	55.21±0.79 ^{cd}
P2	47.07±1.74 ^g	53.17±1.75 ^{def}	57.87±1.21 ^{ab}	59.01±1.24 ^a
P3	47.07±2.41 ^g	52.79±0.91 ^{def}	52.36±1.71 ^{ef}	56.17±0.24 ^{bc}
P4	48.13±1.31 ^g	51.91±0.69 ^f	54.93±1.54 ^{cd}	56.23±1.73 ^{bc}
	pH			
P0	6.05±0.16 ⁱ	6.37±0.06 ^{gh}	6.42±0.12 ^{fgh}	7.20±0.06 ^a
P1	6.03±0.15 ⁱ	6.39±0.16 ^{gh}	6.43±0.11 ^{fg}	7.16±0.13 ^{ab}
P2	6.04±0.12 ⁱ	6.23±0.07 ^h	6.59±0.08 ^{ef}	7.01±0.08 ^{bc}
P3	6.05±0.20 ⁱ	6.48±0.09 ^{fg}	6.51±0.06 ^{fg}	6.86±0.08 ^{cd}
P4	5.95±0.08 ⁱ	6.37±0.07 ^{gh}	6.47±0.06 ^{fg}	6.71±0.20 ^{de}

Note: Different superscripts in the same column and row indicate different results ($P < 0.05$).

CONCLUSION

Eco-enzyme with a composition of rice washing water and yeast (E3) has the best quality and was therefore selected for research. Using eco-enzyme for egg dyeing before storage can maintain egg quality during storage, including yolk and white

quality, and egg weight loss. The shelf life of eggs using eco-enzyme can vary depending on the temperature and environmental conditions used for storage. Higher temperatures and humidity can shorten egg shelf life. In general, treatment P4 is the best treatment for egg dipping.

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