

## **Addition of Tomato Powder in the Diet of Native Laying Hens Formulated with Sardine Oil and Effects on Functional and Sensory Properties of Eggs**

*(Penambahan Tepung Tomat dalam Pakan Ayam Kampung Petelur yang Diformulasikan dengan Minyak Sarden dan Pengaruhnya terhadap Sifat Fungsional dan Sensori Telur)*

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### **ABSTRAK**

Penelitian ini mengevaluasi sifat fungsional dan sensori telur yang diproduksi oleh ayam kampung petelur dengan menggunakan pakan mengandung minyak sarden dan tepung tomat. Sebanyak 120 ayam kampung petelur berumur 28 minggu diberi perlakuan yang berbeda-beda, yaitu kontrol (minyak sarden 0% + tepung tomat 0%), T1 (minyak sarden 2.5% + tepung tomat 0%), T2 (minyak sarden 2.5% + tepung tomat 0,5%), dan T3 (minyak sarden 2.5% + tepung tomat 1%). Masing-masing perlakuan terdiri atas enam ulangan, dan setiap ulangan menggunakan lima ekor ayam. Rancangan Data sifat fungsional dan sensori masing-masing dianalisis menggunakan ANOVA multivariat dan ANOVA satu arah, dan *Duncan's Multiple Range Test* digunakan untuk menunjukkan signifikansi antar perlakuan. Perbedaan perlakuan pakan, waktu penyimpanan, dan suhu penyimpanan menunjukkan interaksi yang signifikan terhadap daya buih ( $P<0,05$ ). Perbedaan perlakuan pakan saja tidak memberikan pengaruh yang signifikan terhadap daya buih. Hasil penelitian juga menunjukkan pengaruh tunggal dari suhu penyimpanan terhadap stabilitas buih ( $P<0,05$ ). Kombinasi tepung tomat dalam pakan tersuplementasi minyak sarden memperbaiki warna, aroma, rasa, dan tekstur kuning telur goreng secara signifikan ( $P<0,05$ ). Penambahan minyak sarden sampai 2,5% dan tepung tomat sampai 1% bertindak secara sinergis dalam meningkatkan sifat sensori tanpa mengurangi sifat fungsional telur. Tepung tomat dapat digunakan dalam formulasi pakan mengandung minyak sarden untuk memberikan pengaruh yang diinginkan pada sifat fungsional dan sensori telur yang dihasilkan oleh ayam kampung petelur.

**Kata kunci:** Daya Buih, Sensori, Stabilitas Emulsi, Telur

### **ABSTRACT**

The functional and sensory properties of eggs from native laying hens fed the diets incorporated with sardine oil and tomato powder were investigated. Native laying hens ( $n=120$ ; 28 weeks old) received four dietary treatments, control (sardine oil 0% + tomato powder 0%), T1 (sardine oil 2.5% + tomato powder 0%), T2 (sardine oil 2.5% + tomato powder 0.5%), and T3 (sardine oil 2.5% + tomato powder 1%). Each treatment comprised six replications and employed five birds each. The experimental design used was a  $3 \times 3 \times 2$  Completely Randomized Factorial Design with the first factor was dietary treatment, the second factor was storage time, and the third factor was storage temperature. Functional and sensory properties data were analyzed by multivariate ANOVA and one-way ANOVA, respectively, and *Duncan's Multiple Range Test* was applied to separate the significant values between treatments. Dietary treatment, storage time, and storage temperature showed a significant interaction with the foaming ability ( $P<0.05$ ). The dietary treatment alone did not give any significant effect on foaming ability. The result also showed a sole significant effect of storage temperature on the foam stability ( $P<0.05$ ). The combination of tomato powder in the sardine oil-supplemented diet improved the color, odor, taste, and texture of fried egg yolk ( $P<0.05$ ). Dietary sardine oil of up to 2.5% and tomato powder of up to 1% acted synergistically in improving sensory properties without decreasing the functional properties of eggs. Tomato powder could be used in feed formulation containing sardine oil to give beneficial effects on the functional and sensory properties of eggs produced by native laying hens.

**Keywords:** Egg, Emulsion Stability, Foaming Ability, Sensory

## INTRODUCTION

Eggs have been widely used in the human diet or as an ingredient in many foods throughout the world. An egg comprises about 29.23% egg yolk, 58.14% albumen, and 12.63% eggshell (Brodacki et al., 2018). Both egg white and egg yolk have their own properties which are believed responsible for changing the taste, texture, and color of food products such as cold souffle, ice cream, and mayonnaise. Ovalbumin, ovotransferrin, ovomucoid, ovomucin, and lysozyme are the functional proteins in egg white associated with some functional properties of egg (Abeyrathne et al., 2013) including foaming capacity and gel formation. Egg yolk through its surface-active components may contribute to the emulsion stability property. These properties can vary quite a bit due to numerous factors such as the rearing system, age of the hens, and feed additive.

This research attention is particularly focused on the use of feed additives because it can be transferred directly into the egg. A previous study by Franchini et al. (2002) found that supplementation of vitamin C and E in the Hy-Line Brown hen's diet improved foam stability and gel elasticity of egg white without lowering the foaming ability, gel hardness, and gel cohesiveness. A recent study conducted by Zhao et al. (2019) demonstrated that the diet supplemented with 450 mg/kg glycerol monolaurate increased the foaming capacity and foaming stability of egg white. It indicates that the diet treatment can determine the desirable qualities of the egg that consequently affect the functional properties of the egg.

Due to the health professional's recommendation to increase the consumption of food sources containing omega-3 fatty acids, egg producers are attempting to produce omega-3 enriched eggs by adding omega-3 sources to the hen's diet. One of the possible approaches is the supplementation of fish oil as a potential source of omega-3 fatty acids in the diet (Eltweri et al., 2016). However, egg yolks resulting from hens fed diets containing fish oil are susceptible to oxidative deterioration during prolonged storage due to high lipid constituents. Lipid oxidation leads to undesirable flavors and free radical compounds. The off-flavors decrease consumer acceptance which is affected by the poor sensory properties of the oxidized egg. There is a further worry considering that the addition of fish oil alone produces fishy-smelling eggs, in particular from the egg yolk. Taking this into account, antioxidant sources from fruits can be used to deodorize fish oil by eliminating the number of volatile compounds, as well as to neutralize free radical compounds.

Tomato contains lycopene, phenolics, vitamin A, vitamin E, and vitamin C, which are well known to be responsible for antioxidant compounds. In addition, to exert the prevention of oxidative damage in egg yolk, as well as improvement of egg quality and fatty acid composition, the tomato powder used in this study was mainly expected to provide the capability of fishy odor masking. A lot of the benefits of tomato indicate that it can be used to reduce the deleterious effect of a diet supplemented by fish oil alone. Therefore, the objective of this study was to evaluate the functional and sensory properties of eggs from native laying hens

fed diets supplemented with sardine oil and tomato powder.

## METHODS

### Laying Hens Rearing and Dietary Treatments

The study was carried out on 120 native laying hens (*Gallus turcicus*) aged 28 weeks old. The hens were housed in individual battery cages (35×45×50 cm; width×depth×height), and they were randomly assigned to four dietary treatments, each treatment consisted of six replications with five birds per replication. The feed given was 100 g/bird for a day and it was administered twice daily, while water was provided *ad libitum*. Dietary treatments consisted of control (sardine oil 0% + tomato powder 0%) (T0), T1 (sardine oil 2.5% + tomato powder 0%), T2 (sardine oil 2.5% + tomato powder 0.5%), and T3 (sardine oil 2.5% + tomato powder 1%). Tomato powder was purchased from the Balai Materia Medica, Batu City. Sardine oil was obtained from the local market in Muncar, Banyuwangi City. The detailed ingredient and nutritional value of dietary treatments have been published in the previous study by Andri et al. (2016a). The trial lasted 6 weeks.

### Experimental Design

Three experimental diets (F1) based on sardine oil and tomato powder were used. An unsupplemented/basal diet was used as a control and other groups were supplemented with 0, 0.5, and 1% of tomato powder while the percentage (2.5%) of sardine oil remained the same. Three experimental storage times (F2) were arranged on days 7, 14, and 21. Two experimental storage temperatures (F3) were set at 4 and 25°C.

### Egg Functional Properties

After 6 weeks of dietary treatment, forty eggs from each dietary treatment were collected for this analysis. They were then stored at room temperature (25°C) and another set of eggs was kept under refrigeration (4°C), and subjected to different storage periods (7, 14, and 21 days). Foaming ability, foam stability, and emulsion stability were measured according to the following methods.

About 50 mL of egg white was whipped using a mixer for 5 minutes, in which a medium speed was applied for 2.5 minutes at the beginning and the remaining time for the higher speed. Foaming ability was calculated as follows (Chalamaiah et al., 2010):

$$\text{Foaming ability (\%)} = \left[ \frac{a-b}{b} \right] \times 100$$

Where: a = total volume after whipping; b = initial volume

The foam obtained after whipping was then allowed to drain for an hour at room temperature. The amount of foam stability in terms of foam drainage was calculated using the following expression (Yang and Baldwin, 1995):

$$\text{Foam stability (\%)} = \left( \frac{a}{b} \right) \times 100$$

Where: a = volume of solution drained from foam; b = volume of foam

Measurement of emulsion stability was done by following the explanation of Harrison and Cunningham (1986) with modifications in the ingredients used and the condition of centrifugation. The ingredients used were 16 g of egg yolk, 2 mL of coconut oil, and 10 mL of apple cider vinegar. The mixture was whipped for 5 minutes and was placed in a sealed glass test tube and then stored at an ambient

temperature for 48 hours. The emulsion was separated using a centrifuge at the condition of 3000 rpm for 20 minutes after completing the storage period. The milliliters of separated oil was recorded as emulsion stability (mL).

### **Sensory Evaluation**

In this evaluation, twenty-five untrained panelists of the Faculty of Animal Science, Universitas Brawijaya, were recruited to evaluate the boiled and fried eggs. Each panelist received two cooking types (boiled and fried) of eggs, and each plate consisted of eight samples (four egg yolks and four egg whites). The boiled and fried eggs were made without any additional ingredients, and the eggs were fried without cooking oil. Attributes such as color, odor, texture, taste, and overall acceptability were instructed to the panelist and used a 5-point hedonic scale (5=like very much; 4=like somewhat, 3=neither like nor dislike, 2=dislike somewhat, 1=dislike very much).

### **Statistical Analysis**

A multivariate analysis of variance (MANOVA) was run using SPSS 16.0 to analyze data on functional properties. Foaming ability, foam stability, and emulsion stability were set as dependent variables, while dietary treatment, storage time, and storage temperature were assigned as fixed factor(s). Data of sensory properties were statistically analyzed by one-way analysis of variance (ANOVA) using SPSS 16.0. When significant effects were found, mean values were separated by using Duncan's multiple range test with a confidence level of 95%.

## **RESULTS AND DISCUSSION**

### **Foaming Ability and Foam Stability**

The result shows that there was an interaction ( $P<0.05$ ) between dietary treatment, storage time, and storage temperature on the foaming ability (Table 1). Regarding storage time levels, the highest value for foaming ability was observed at 21 days of storage ( $P<0.05$ ). A work by Biladeau and Keener (2009) was in line with this study and demonstrated the highest egg foam volume of the control treatment (9.16 mL/g of albumen) was recorded at the longest storage time (12 weeks storage). The shorter storage time (7 and 14 days) resulted in lower foaming ability, 421.88% and 453.12% ( $P>0.05$ ), respectively. Previous studies also reported the foaming ability of egg white stored for up to 1 month remained stable due to the pH values of egg whites were also relatively constant during storage (Liu et al., 2009; Kralik et al., 2021). Hence, Kenawi et al. (2016) revealed the foaming ability was greatly influenced by the storage temperature rather than the storage time. Even if they are stored for a prolonged time, the egg whites will be maintained in cold storage conditions.

In this study, the foaming ability value increased ( $P<0.05$ ) as the storage temperature of the egg elevated (Table 1). Altalhi (2013) also found a similar result in which a higher temperature of egg white (20°C) resulted in higher foaming ability (724%), whereas a lower temperature (4°C) only obtained 682%. A considerable rise can be mainly caused by the pH of egg white that increases due to the loss of carbon dioxide through the pores in the eggshell during storage under high temperatures. Cold storage at 4°C maintained lower pH (8.91 to 8.99 from day 7 to 21) compared with the other egg whites stored at room temperature (9.05 to

9.15) (data not shown). At higher pH, the foaming ability is also found to be higher due to the increase of protein flexibility caused by the enhancement of net charges on the protein (Khalid and Elhardallou, 2015). The formation and stability of the foam can be related to the interaction between positively charged lysozyme and other negatively charged proteins.

Result also demonstrates that the foam stability was only affected by the storage temperature ( $P < 0.05$ ) (Table 1), and it indicates that the egg white pH possibly influences the foam. In fact, the foam stability increased with increasing the storage time, in this agreement with those

found by Liu et al. (2009), who observed the foam stability became higher after 5 days of storage ( $P > 0.05$ ). Foam destabilization can happen in higher pH and extended storage due to the protein denaturation, as well as rupture and coalescence of the air bubbles. However, it does not happen in this study. A lot of factors may modify the functional properties of egg white such as whipping time and speed (Altalhi, 2013), egg yolk contamination (Yao et al., 2014), egg treatments such as irradiation and ultrasonic (Liu et al., 2009), as well as dietary treatments (Zhao et al., 2019).

Table 1. Functional properties of egg as affected by dietary treatments, storage time, and storage temperature.

Parameters	Foaming ability (%)	Foam stability (%)	Emulsion stability (mL)
-----F1 = Dietary treatments-----			
Control	491.67±116.45	40.67±9.88	0.65±0.17
T1	466.67±109.40	40.33±5.18	0.64±0.17
T2	445.83±135.61	43.83±8.29	0.67±0.15
T3	462.50±102.53	37.50±7.78	0.74±0.14
-----F2 = Storage time (day) -----			
7	421.88±79.52 a	38.50±8.18	0.52±0.11 a
14	453.12±120.37 a	39.25±6.92	0.70±0.10 b
21	525.00±118.32 b	44.00±8.26	0.79±0.11 c
-----F3 = Storage temperature (°C) -----			
4	416.67±114.84 a	38.17±6.54 a	0.65±0.18
25	516.67±90.50 b	43.00±8.77 b	0.70±0.14
-----P-value-----			
F1	ns	ns	ns
F2	*	ns	*
F3	*	*	ns
F1 x F2	*	ns	ns
F1 x F3	ns	ns	ns
F2 x F3	ns	ns	ns
F1 x F2 x F3	*	ns	ns

a,b,c Values (mean±standard deviation) with different superscript letters in the same columns are significantly different ( $P < 0.05$ ); \* is significant at the level of  $P < 0.05$ , ns: not significant ( $P > 0.05$ ); Control (sardine oil 0% + tomato powder 0%); T1 (sardine oil 2.5% + tomato powder 0%); T2 (sardine oil 2.5% + tomato powder 0.5%); T3 (sardine oil 2.5% + tomato powder 1%)

This study indicates that dietary treatments likely affected foam stability even though there was no significant

difference ( $P > 0.05$ ) (Table 1). Diet supplemented with sardine oil 2.5% and tomato powder 0.5% (T2) showed the

highest foam stability. This means that the composition of egg white changed, in particular the lipid content of egg white. The very early study by Pankey and Stadelman (1969) confirmed that the lipid content of egg whites from hens fed the diet supplemented with each soybean oil, hydrogenated coconut oil, safflower oil, and olive oil slightly increased from 0.0017% to 0.0022% ( $P>0.05$ ). In other words, dietary treatment may increase the dry matter and lower the water content of egg white, and eventually prevent fluid loss by water drainage. This study also highlights the effects of storage time and storage temperature were much higher than dietary treatment.

### **Emulsion Stability**

Storage time decreased the emulsion stability formed by egg yolk ( $P<0.05$ ), which was indicated by the increase in the volume of oil separated during storage (Table 1). Biladeau and Keener (2009) and Karshenas et al. (2018) showed a similar pattern, in which emulsion stability decreased during prolonged storage and the end of storage resulted in the highest separation of oil. It is suspected that vitelline membrane strength weakens over time due to protein degradation and finally disintegrates the fiber network of the vitelline membrane (Biladeau and Keener, 2009). It should be remembered that the egg white pH increases as a function of storage and converts thick egg white to thin egg white. This is possibly transferred into the yolk and modifies the egg yolk pH. The increasing egg yolk pH and egg white pH are mostly correlated with increasing storage time.

Egg yolk itself is a kind of oil-in-water emulsion, while the protein and

lipoprotein contents in the egg form a film that layers around the drops of oil to prevent coalescence with each other of an emulsion system (Karshenas et al., 2018). Considering egg yolk composition, the dietary treatment may exert an effect on the emulsion stability of egg yolk through the changes in the fatty acid composition. Andri et al. (2016b) reported that the addition of fish oil and tomato powder greatly improved the omega-3 fatty acid content of chicken eggs, two times higher than the control group. A study by Kralik et al. (2020) also showed that the compositions of chicken eggs rich in omega-3 fatty acids were completely changed in the saturated fatty acid, monounsaturated fatty acid, and polyunsaturated fatty acid levels due to the feeding of a fish oil-based diet. They also found that TBARS values of fresh and stored eggs were significantly higher since high unsaturated fatty acids induced an oxidation process owing to their double bond structures.

As shown in Table 1, dietary treatment with sardine oil lowered the emulsion stability of egg yolk ( $P>0.05$ ) which was indicated by the higher volume of separated oil. This can be due to rapid oxidation that degrades fatty acid structures and weakens the yolk strength, so the emulsion of egg yolk becomes unstable, too. Emphasizing this matter, the author suggests encapsulating the sardine oil and tomato powder before incorporating them into the animal diet in order to protect these bioactive compounds against physical and chemical degradations.

### **Sensory Properties**

Sensory evaluation scores of attributes of fried- and boiled- egg yolks

are given in Table 2. These results show that supplementation of sardine oil and tomato powder significantly ( $P < 0.05$ ) improved the color of fried egg yolk. This can be attributed to the content of sardine oil and or tomato powder. As reported by Rotolo et al. (2010) that diet supplemented with lycopene, a red pigment, from tomato extraction could be deposited in the egg yolk and contributed to its color intensity. In addition, Saleh (2013) observed a darker egg yolk color (9.16) was obtained at a supplementation dose of 5% fish oil. Perhaps both materials can interact with egg yolk lipids because they belong to fat-

soluble compounds and consequently intensify the egg yolk color.

Also, the interaction significantly ( $P < 0.05$ ) increased the texture of fried- and boiled- egg yolk (Table 2). Confirming our result, Sujatha and Narahari (2011) also found that the egg yolk texture was enhanced from 3.0 to 3.7 (4-point hedonic scale) due to the diet treated with the combination of fish oil, flaxseed, vitamin E, organic Se, and spirulina. This result supposes that egg yolk composition plays an important role to modify the egg yolk texture.

Table 2. Sensory properties of fried and boiled egg yolks as affected by dietary treatments.

Type	Attributes	Dietary treatments				P-value
		Control	T1	T2	T3	
Fried	Color	3.79±0.29 <sup>a</sup>	4.20±0.25 <sup>b</sup>	4.25±0.42 <sup>b</sup>	4.50±0.22 <sup>b</sup>	*
	Odor	3.50±0.45 <sup>d</sup>	1.88±0.34 <sup>a</sup>	2.50±0.35 <sup>b</sup>	3.00±0.27 <sup>c</sup>	*
	Taste	3.04±0.10 <sup>a</sup>	3.67±0.63 <sup>b</sup>	3.38±0.44 <sup>ab</sup>	2.96±0.33 <sup>a</sup>	*
	Texture	2.75±0.22 <sup>a</sup>	3.38±0.49 <sup>b</sup>	3.17±0.20 <sup>ab</sup>	3.63±0.56 <sup>b</sup>	*
	Overall	3.00±0.10	3.17±0.63	3.38±0.34	3.67±0.38	ns
Boiled	Color	3.88±0.26	3.80±0.29	3.92±0.44	4.00±0.52	ns
	Odor	3.67±0.40	3.00±0.80	3.25±0.55	3.33±0.72	ns
	Taste	2.88±0.30 <sup>a</sup>	4.04±0.33 <sup>c</sup>	3.30±0.33 <sup>b</sup>	3.38±0.26 <sup>b</sup>	*
	Texture	3.00±0.45 <sup>a</sup>	3.38±0.34 <sup>a</sup>	3.30±0.30 <sup>a</sup>	3.92±0.40 <sup>b</sup>	*
	Overall	3.30±0.25	3.13±0.63	3.33±0.38	3.42±0.50	ns

a,b,c Values (mean±standard deviation) with different superscript letters in the same rows are significantly different ( $P < 0.05$ ); \*significant at the level of  $P < 0.05$ , ns: not significant ( $P > 0.05$ ); Control (sardine oil 0% + tomato powder 0%); T1 (sardine oil 2.5% + tomato powder 0%); T2 (sardine oil 2.5% + tomato powder 0.5%); T3 (sardine oil 2.5% + tomato powder 1%)

Table 3. Sensory properties of fried and boiled egg whites as affected by dietary treatments.

Type	Attributes	Dietary treatments				P-value
		Control	T1	T2	T3	
Fried	Color	2.83±0.38	3.08±0.44	3.00±0.45	3.13±0.38	ns
	Odor	4.38±0.30	4.04±0.40	4.00±0.16	4.13±0.20	ns
	Taste	2.92±0.30	3.25±0.57	3.25±0.27	3.04±0.29	ns
	Texture	3.33±0.50	3.30±0.37	3.42±0.66	3.46±0.40	ns
	Overall	4.20±0.40	4.08±0.40	4.00±0.52	4.13±0.26	ns
Boiled	Color	4.08±0.30	3.96±0.51	3.88±0.30	3.92±0.34	ns
	Odor	4.20±0.40	4.08±0.34	4.00±0.32	3.96±0.25	ns
	Taste	3.00±0.32	3.33±0.13	3.17±0.38	3.17±0.26	ns
	Texture	3.54±0.19	3.67±0.30	3.70±0.48	3.67±0.34	ns
	Overall	3.38±0.40	4.08±0.40	4.00±0.52	4.13±0.26	ns

ns : not significant ( $P > 0.05$ ); Control (sardine oil 0% + tomato powder 0%); T1 (sardine oil 2.5% + tomato powder 0%); T2 (sardine oil 2.5% + tomato powder 0.5%); T3 (sardine oil 2.5% + tomato powder 1%)

The same effect was also observed in the taste of fried- and boiled- egg yolks (Table 2). Regardless of cooking type, the result demonstrates the significant contribution ( $P < 0.05$ ) of diets treated with sardine oil alone or in combination with tomato powder to produce a better egg yolk taste than the control group. Although omega 3-enriched eggs are commonly associated with a fishy taste (Fraeye et al., 2012), the result of the present study suggests that the levels of sardine oil used can still produce an acceptable egg organoleptically.

Another effect was also found in the significant reduction ( $P < 0.05$ ) of odor acceptance of fried egg yolk supplemented with sardine oil alone (T1). Even so, the addition of tomato powder at concentrations of 0.5% and 1% could significantly improve odor acceptance of fried egg yolk ( $P < 0.05$ ), but insignificant to the odor of boiled egg yolk ( $P > 0.05$ ). With respect to volatile compounds, their availability can be abundant due to the thermo-oxidative degradation of fats during the frying process that leads to the formation of off-odors (Brühl, 2014). Furthermore, the antioxidant effect of tomato powder promotes the inhibition of volatiles formation, thus attenuating the off-odors. Meanwhile, dietary treatments did not affect the sensory properties of fried- and boiled- egg white (Table 3). Nonetheless, our present study is the first contribution to the understanding of sensory and functional properties of eggs produced by hens fed diets supplemented with sardine oil without or with tomato powder.

#### **CONCLUSIONS AND RECOMMENDATIONS**

To conclude, the results of this study indicate that the addition of tomato powder and sardine oil in the diet of native laying hens could have no effect on the functional properties of eggs but a higher concentration of tomato powder had a

positive effect on the sensory properties of eggs.

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