Quality of Japanese quail eggs according to different storage periods and temperatures

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**ABSTRACT.** The objective was to evaluate the quality of Japanese quail eggs stored at room temperature (26.8°C) and under refrigeration (10.9°C), for different storage periods (0; 5; 10; 15; 20; 25 and 30 days). A total of 196 quail eggs were assigned to different treatments in a factorial arrangement of 2 (temperatures) x 7 (storage periods), with 3 replications and 4 eggs per experimental unit. Data were subjected to regression analysis of parameters as a function of storage time at room temperature and under refrigeration. There was a reduction in egg weight, albumen weight and height, yolk height and egg yolk index with increasing storage period as a function of temperatures (p < 0.05). Eggs at room temperature showed a reduction in albumen index, while eggs under refrigeration showed a quadratic behavior (p < 0.05). Storage days promoted a quadratic response in albumen and yolk pH (p < 0.05) of eggs at room temperature. Eggs at room temperature floated from the 15th day of storage for up to 30 days. Refrigerated storage is recommended.

Keywords: yolk height; coturniculture; product of animal origin; shelf life.

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### Introduction

Coturniculture has shown a marked growth in recent years, seeking to adapt to new production technologies, ceasing to be a subsistence activity and occupying a highly modern and productive scenario, with promising results. Japanese quail (*Coturnix coturnix japonica*) is a poultry species commercially bred for meat and egg production (Taha, El-Tahawy, El-Hack, Swelum, & Saadeldin, 2019), and is also used for experimental purposes (Grzegrzółka & Gruszczyńska, 2019). Large-scale production of quail eggs and meat is feasible, in view of the high reproductive potential, rapid growth rate, high feed conversion capacity, sexual precocity, short generation interval, small space requirement, low investment. and resistance to heat and diseases, which makes quail farming an excellent alternative for the production of animal protein (Furtado, Braz, Nascimento, Lopes Neto, & Oliveira, 2018; Taha, El-Tahawy, El-Hack, Swelum, & Saadeldin, 2019).

Although chicken eggs dominate the egg market, in countries such as China, Japan, Brazil and France, Japanese quail eggs have a significant share in the economy (Ondrušíková, Nedomová, Pytel, Cwiková, & Kumbár, 2018), with production of eggs estimated at 200–250 eggs year<sup>-1</sup> (Warsito et al., 2021). The popularity of quail eggs is because they are considered a complete and balanced food in nutrients, meeting the criteria of a food with functional properties (Adamski, Kuźniacka, Czarnecki, Kucharska-Gaca, & Kowalska, 2017). Despite their small size (10 - 12 g), quail eggs are rich in proteins, amino acids, vitamins and minerals and have low levels of triglycerides and saturated fatty acids (Ondrušíková, Nedomová, Pytel, Cwiková, & Kumbár, 2018). In addition, quail egg protein is considered hypoallergenic, which makes it an alternative for people allergic to chicken egg protein (Nowaczewski et al., 2021)

However, quail eggs are perishable and can lose their internal quality after laying if not properly stored. Thus, for the full nutritive potential of the egg to be used and to have a longer shelf life, it is necessary to store the quail egg under an ideal temperature and for an adequate period (Ibrahim, Abare, Salisu, & Abdulkarim, 2020). Regarding that the binomial time and temperature directly affect the quality of eggs, and must be controlled to extend the shelf life (Yimenu, Kim, Koo, & Kim, 2017). Thus, Silva et al. (2020) mention that for proper storage, quail eggs must be stored at a temperature ranging from 10 to 15°C and relative humidity between 70 and 80%.

Long storage periods reduce the internal quality of eggs. With prolonged storage, egg weight loss rates increase, albumen becomes less viscous due to chemical reactions related to moisture and  $CO_2$  loss, causing height reduction, alkalinity increase and palatability loss. According to Akpınar and Günenç (2019), quail eggs can preserve quality attributes at room temperature for storage periods of less than four days. Thus, we hypothesized that the storage of quail eggs under refrigeration reduces the deterioration rate and the eggs can be stored for periods of up to 30 days, under these conditions, without losing internal quality.

Due to limited information on the storage conditions of quail eggs, the recommendations used for their storage are those intended for chicken eggs (Renukadevi, Himali, & Silva, 2018). Thus, studies on the interaction storage period x temperature of quail eggs are necessary and valid, so that high quality products are available to the consumer market.

Thus, the objective was to evaluate the internal quality of Japanese quail eggs, subjected to different storage periods and temperatures.

# Material and methods

### **Experiment location**

The experiment was carried out at the Poultry Laboratory of the Federal University of São Francisco Valley (UNIVASF), Petrolina, state of Pernambuco, Brazil (9°19'28" South latitude, 40°33'34" West longitude, 393m altitude). The climate is hot semi-arid (Köppen & Geiger 1928), with a rainy season (BSh), with 376 mm average annual rainfall, 26°C average annual temperature and approximately 61% average relative humidity.

#### Eggs

A total of 196 eggs, with 10.5 g average weight, from Japanese quails (*Coturnix coturnix japonica*), with 27 weeks of age and coming from the same rearing system, were selected for no deformation and/or cracks, and were subsequently sanitized, identified and packed in commercial plastic packaging (capacity for 30 eggs) according to the treatments.

### Treatments

Eggs were distributed in a completely randomized design, in a 2 (temperatures) x 7 (storage periods) factorial arrangement, totaling 14 treatments, with 3 replications of 4 eggs per experimental unit. The treatments consisted of two storage conditions: room temperature ( $26.8^{\circ}C \pm 1.0^{\circ}C$  and 55% relative humidity) and under refrigeration (10.9°C and 63% relative humidity). Eggs were analyzed during a period of 30 days, with evaluations carried out in different storage periods (0, 5, 10, 15, 20, 25 and 30 days).

Temperature and relative humidity were recorded every two days, within each storage condition (at room temperature and under refrigeration), throughout the experimental period, using digital thermo-hygrometers (Incoterm, 7664.01.0.00, São Paulo, São Paulo State, Brasil) distributed in the storage room at room temperature and inside the refrigerator (Electrolux, DC35A, Rio de Janeiro, Rio de Janeiro State, Brasil).

# Egg weight

Eggs were individually weighed on an analytical balance (Tecnal, SHI-BL-3200H, Piracicaba, São Paulo State, Brasil) on the first experimental day (day zero), and then stored under specific conditions (room temperature and under refrigeration). At the end of each storage period, eggs were weighed again (Oliveira et al., 2021).

#### Floating

For float test, two eggs from each treatment were separated and immersed in 500 mL fresh water for 3 minutes. After, it was evaluated whether the egg floated (+) or not (-).

### Shell, yolk and albumen weight

Eggs were broken on a flat, smooth polyethylene surface and the yolks were separated from the albumen. Albumen and yolk were separated using a 20 mm syringe. A syringe was used for each treatment, which was washed with distilled water according to each egg to be analyzed.

Shells were carefully washed and dried at room temperature for 48h and then weighed. The separation of egg components is illustrated in Figure 1.



Figure 1. Separation of egg components.

The percentages of shell, yolk and albumen were determined by the equations (Nemati et al., 2020):

% shell = (shell weight/ egg weight) \* 100

% yolk = (yolk weight/egg weight) \* 100

% albumen= 100 - (% yolk + % shell)

# Albumen and yolk measurements and indices

Albumen and yolk height and diameter (in mm) were measured with a digital caliper (FG8331, Franca, São Paulo State, Brazil). Data obtained in the measurements were used to determine the indices (Ondrušíková, Nedomová, Pytel, Cwiková, & Kumbár, 2018):

Albumen index (%): albumen height / albumen diameter \* 100

Yolk index (%): yolk height / yolk diameter \* 100

# pH determination

Values of pH were measured in the albumen and yolk of each egg using a benchtop pH meter (Tecnal, R-TEC-7-MP, Piracicaba, São Paulo State, Brazil), previously calibrated with pH 4.0 and 7.0 buffer solutions. The albumen and the yolk were deposited in recipients for further dilution in distilled water (100 mL) followed by homogenization. The samples were left to rest following the recommendations of the AOAC (2016). The pH was read by immersing the probe into each recipient with the samples.

# Statistical analysis

Data were analyzed using PROC GLM of the Statistical Analysis System University Software (SAS University) by means of analysis of variance and regression of parameters as a function of storage time at room temperature and under refrigeration. The significance of the parameters estimated by the models and the coefficients of determination were adopted as criteria for choosing the regression models. To estimate the regression equation between pressure and volume data, the PROC REG procedure was used. The following statistical model was used:

 $Y = \mu + Tj + eij$ 

where:  $\mu$  = overall mean; Tj = days of storage as a function of temperature; eij = residual error.

# **Results and discussion**

Based on the results obtained, the weight of the tested quail eggs decreased linearly as a function of room (p < 0.001) and refrigerated (p = 0.0015) temperatures during the 30 days of storage (Table 1).

Temperature			S	torage da			P-value				
	0	5	10	15	20	25	30	Mean	SEM	L	Q
			Eg	g weight	(g)						
A (26.8°C)	10.7	10.5	10.3	9.7	9.9	9.5	9.6	10.0	0.08	<0.001 <sup>a</sup>	0.3435
R (10.9°C)	10.3	11.2	10.6	9.9	10.3	10.3	9.7	10.3	0.08	$0.0015^{b}$	0.2015
			She	ell weight	: (g)						
A (26.8°C)	0.84	0.86	0.93	0.85	0.90	0.92	0.90	0.89	0.01	0.091	0.5083
R (10.9°C)	0.86	0.94	0.90	0.87	0.92	0.89	0.85	0.89	0.01	0.4467	0.0995
			Albu	men weig	ht (g)						
A (26.8°C)	6.10	5.84	5.59	4.96	5.12	4.77	4.77	5.31	0.13	<0.001 <sup>c</sup>	0.1424
R (10.9°C)	5.88	6.35	5.88	5.51	5.71	5.58	4.98	5.70	0.13	<0.001 <sup>d</sup>	0.1135
			Yo	lk weight	(g)						
A (26.8°C)	3.71	3.77	3.77	3.89	3.86	3.80	3.94	3.82	0.12	0.2526	0.8783
R (10.9°C)	3.56	3.95	3.84	3.54	3.67	3.82	3.87	3.75	0.12	0.5188	0.7246

Table 1. Weight of components of quail eggs stored at room temperature (26.8°C) and under refrigeration (10.9°C) for 30 days of storage.

A = Room temperature; R = Refrigeration temperature; SEM = standard error of the mean; L = significant for linear effect; Q = significant for quadratic effect. Significant at the 5% probability level. Equations:  $\hat{Y}^a = 10.6834 - 0.0337x$ ;  $R^2 = 0.72$ ;  $\hat{Y}^b = 10.7640 - 0.0283x$ ;  $R^2 = 0.39$ ;  $\hat{Y}^c = 6.0869 - 0.0385x$ ;  $R^2 = 0.88$ ;  $\hat{Y}^d = 6.1679 - 0.03128x$ ;  $R^2 = 0.65$ .

Eggs stored at room temperature showed lower weight in relation to eggs stored under refrigeration, reducing their weight throughout the storage period, from 10.7 g (day 0) to 9.6 g on the thirtieth day of storage at room temperature. This reduction in egg weight is related to the respiratory activity of the egg, which releases water vapor, CO<sub>2</sub>, ammonia, nitrogen and hydrogen sulfide gas to the external environment (Dada, Raji, Akinoso, & Aruna, 2018). This process begins soon after laying, and can be accelerated by improper storage conditions, especially when eggs are subjected to high temperatures, given that egg packaging at room temperature promotes faster dehydration of the cuticle that plug the eggshell pores, which undergoes shrinkage, thus allowing the shell pores to increase in size, which facilitates the evaporation of gases and water from the albumen (Oliveira et al., 2021).

The linear reduction in albumen weight of quail eggs as a function of room (p < 0.001) and refrigerated (p < 0.001) (Table 1) temperatures directly affected albumen height (p < 0.05; Table 2), which was also reduced with increasing storage days, as a function of storage temperatures. Eggs stored at room temperature had lower albumen weight (5.31 g; Table 1) and height (2.81 mm; Table 2) compared to eggs kept under refrigeration during a period of 30 days of storage. The reduction in albumen height during the 30 days of storage at room temperature (Table 2) possibly occurred due to denaturation of ovomucin, which resulted in the release of water that was associated with this protein, which makes the albumen more fluid by reducing its viscosity, leaving it more spread out (Wang, Wang, & Shan, 2019). Our results corroborate the findings of Nemati et al. (2020), who observed a greater reduction in albumen height of quail eggs stored at room temperature ( $22^{\circ}$ C) for 30 days of storage compared to eggs stored in a refrigerated environment ( $5^{\circ}$ C). There was no effect of storage period as a function of temperatures on the quail eggshell weight (p > 0.05; Table 1).

The breakdown of carbonic acid makes the albumen more fluid. At this moment, it crosses the vitelline membrane by osmosis and reaches the yolk, which absorbs water from the albumen in an attempt to equalize the concentration (pressure) between the two phases (albumen and yolk), which leads yolk swelling, which in turn exerts pressure on the vitelline membrane. This pressure causes the yolk to change from a spheroid shape to a flaccid, flat shape, reducing its height (Eke, Olaitan, & Ochefu, 2013; Nasri, van den Brand, Najjar, & Bouzouai, 2020), in addition to easy breakage of the yolk during egg handling. In our study, although yolk weight was not influenced by storage days as a function of temperatures (p > 0.05; Table 1), the yolk height linearly reduced during the storage period for both eggs stored at room temperature (p < 0.001; Table 2) and for eggs stored under refrigeration (p = 0.0038; Table 2), with a greater decrease in yolk height of quail eggs stored at room temperature, from 11.04 mm (day 0) to 4.24 mm at the end of the storage period. For eggs subjected to refrigeration, the reduction in yolk height occurred more slowly, in which the eggs at the end of the storage period showed a mean yolk height of 9.00 mm, indicating that eggs stored in a refrigerated environment showed better preservation compared to those stored at room temperature.

One of the most important qualities of quail eggs is the albumen index, which directly correlates with their height and width. According to Hassan, Mohammed, Hussein, and Hussen (2017), fresh eggs have a higher albumen index than older eggs. Following this premise, it appears that the mean values of the albumen index of the quail eggs evaluated in the present study were linearly reduced throughout the storage period at room temperature, ranging from 0.080 (day 0) to 0.025 (day 30) (p < 0.001; Table 2), with the lowest mean albumen

index verified for this storage condition (0.049: Table 2), in relation to quail eggs stored under refrigeration. For Dada, Raji, Akinoso, and Aruna (2018), storage temperatures above 18°C favor metabolic activities within the egg, which lead to greater movement of water from the albumen to the yolk, reducing the albumen quality.

**Table 2.** Mean values of height, index and pH of albumen and yolk of quail eggs during the period of 30 days of storage at room(26.8°C) and refrigeration (10.9°C) temperatures.

	Storage days								CEM	P-value	
Temperature	0	5	10	15	20	25	30	Mean	SEM	L	Q
			Albun	nen height	t (mm)						
A (26.8°C)	3.62	3.85	2.77	2.77	2.50	2.10	2.06	2.81	0.21	<0.001 <sup>a</sup>	0.582
R (10.9°C)	3.91	3.90	2.99	3.05	3.38	3.04	2.64	3.27	0.21	$0.0023^{b}$	0.558
			Yoll	k height (1	mm)						
A (26.8°C)	11.04	9.33	6.61	6.16	5.30	4.64	4.24	6.76	0.34	<0.001 <sup>c</sup>	0.000
R (10.9°C)	9.70	10.50	9.82	8.99	8.87	8.08	9.00	9.28	0.40	$0.0038^{d}$	0.714
			Albu	men inde	x (%)						
A (26.8°C)	0.080	0.080	0.050	0.039	0.032	0.033	0.025	0.049	0.003	<0.001 <sup>e</sup>	0.030
R (10.9°C)	0.090	0.080	0.063	0.053	0.054	0.055	0.048	0.063	0.003	< 0.001	0.005
			Yc	olk index (	%)						
A (26.8°C)	0.395	0.338	0.216	0.194	0.153	0.141	0.128	0.224	0.01	<0.001 <sup>g</sup>	0.000
R (10.9°C)	0.399	0.381	0.367	0.341	0.322	0.337	0.305	0.350	0.07	<0.001 <sup>h</sup>	0.205
			А	lbumen p	Н						
A (26.8°C)	8.69	8.60	9.39	9.44	9.51	9.46	9.10	9.17	0.14	0.0078	0.006
R (10.9°C)	9.58	9.24	9.48	9.38	9.36	9.32	9.31	9.38	0.03	0.3301	0.265
				Yolk pH							
A (26.8°C)	6.05	6.26	6.81	7.99	7.35	7.79	7.29	7.08	0.18	< 0.001	0.003
R (10.9°C)	6.58	6.52	6.74	6.67	6.71	6.66	6.53	6.63	0.10	0.876	0.159

A = Room temperature; R = Refrigeration temperature; SEM = standard error of the mean; L = significant for linear effect; Q = significant for quadratic effect. Significant at the 5% probability level. Equations:  $\hat{Y}^a = 3.7707 - 0.0486x$ ;  $R^2 = 0.84$ ;  $\hat{Y}^b = 3.8251 - 0.0368x$ ;  $R^2 = 0.68$ ;  $\hat{Y}^c = 10.0933 - 0.2221x$ ;  $R^2 = 0.90$ ;  $\hat{Y}^d = 10.1240 - 0.0561x$ ;  $R^2 = 0.59$ ;  $\hat{Y}^e = 7.9035 - 2.0214x$ ;  $R^2 = 0.93$ ;  $\hat{Y}^r = 0.0877 - 0.0034x + 0.00006x^2$ ;  $R^2 = 0.94$ ;  $\hat{Y}^s = 0.36126 - 0.0091x$ ;  $R^2 = 0.94$ ;  $\hat{Y}^h = 0.3896 - 0.0026x$ ;  $R^2 = 0.71$ ;  $\hat{Y}^i = 8.5117 + 0.1007x - 0.0026x^2$ ;  $R^2 = 0.79$ ;  $\hat{Y}^i = 5.8325 + 0.1627x - 0.0037x^2$ ;  $R^2 = 0.81$ .

A quadratic effect was found for the albumen index of quail eggs stored under refrigeration (p = 0.0059; Table 2), with reduction in albumen index until the 15<sup>th</sup> day of storage, rising during the 20 and 25<sup>th</sup> days and returning to reduce from the 28<sup>th</sup> day of storage onwards. This mechanism can be explained by the increase in the albumen axis, which consequently results in a decrease in the index, since the index is a variable dependent on the axis and height of the albumen.

As the egg deteriorates, the yolk index decreases due to the breakdown of the fibrous glycoprotein ovomucin (Muhammad et al., 2016). In the present study, the yolk index, similar to the albumen index, showed a decreasing linear behavior as a function of room (p < 0.001; Table 2) and refrigerated (p < 0.001; Table 2) temperatures as the storage days increased, with lower yolk indices for eggs stored at room temperature.

The yolk indices of the evaluated quail eggs showed mean values of 0.224 for eggs at room temperature and 0.350 for eggs under refrigeration (Table 2). Taking into account the yolk index values presented by Eke, Olaitan, and Ochefu (2013) for classification of fresh eggs (0.30-0.50), it can be inferred that the yolks of quail eggs stored in a refrigerated environment presented better quality than those stored at room temperature. Yolk index values lower than 0.25 indicate a very fragile yolk (Qi, Zhao, Li, Shen, & Lu, 2020).

The lowest yolk index observed for quail eggs stored at room temperature is possibly related to the lower mean height (6.76 mm) that yolks of these eggs presented in relation to eggs kept under refrigeration (9.28 mm) (Table 2). According to Santos, Segura, and Sarmiento (2019), high temperatures cause stretching and increased permeability of the vitelline membrane, which accelerates the transfer of water from the albumen to the yolk, which has higher osmotic pressure, causing it to lose its spherical shape and becoming more elongated and flattened, causing a reduction in the yolk index.

During the storage period of eggs at room temperature, the albumen pH showed a quadratic behavior (p = 0.0060; Table 2), in which the eggs initially had a mean pH of 8.69 (day 0), reaching the highest albumen pH value on the 20th day of storage, declining in sequence, presenting at the end of the experimental period a final pH of 9.10 (day 30). Probably the increase in pH of eggs kept at room temperature was caused by the greater loss of carbon dioxide from the egg through shell pores and thus increased albumen alkalinity (Kumari, Tripathi, Maurya, & Kumar, 2020). It can also be attributed to the high-temperature catalytic effect

of the enzyme carbonic anhydrase, which dissociated H<sub>2</sub>CO<sub>3</sub> into H<sub>2</sub>O and CO<sub>2</sub> and these evaporated through the shell pores, thus increasing the albumen pH (Dada, Raji, Akinoso, & Aruna, 2018).

The lowest mean pH values recorded in this study were measured on days 0 (8.69) and 5 (8.60) of storage at room temperature (Table 2). These values are below the pH limit (9.0-9.7) considered adequate for eggs subjected to different storage periods (Qi, Zhao, Li, Shen, & Lu, 2020). There was no effect of the storage period on the pH of quail eggs kept under refrigeration (p > 0.05; Table 2).

For yolk pH, a quadratic behavior was verified for eggs stored at room temperature (p = 0.0031; Table 2), with higher yolk pH at 15 days of storage (7.99; Table 2), with all yolk pH values from this period above the range 6.0-6.9 recommended by Dutra et al. (2021). Considering that in eggs stored at room temperature, the transfer from albumen to the yolk is more pronounced, the increase in yolk pH possibly occurred due to alkaline ions from the albumen that can be exchanged with H+ ions present in the yolk, resulting in an increase in the yolk pH (Feddern et al., 2017).

Although no significant effect was detected on yolk pH of eggs subjected to refrigeration (p > 0.05; Table 2), the yolk pH values of these eggs (6.52 - 6.74) were within the limits established by Dutra et al. (2021) for good quality eggs.

Regarding the floating of quail eggs, eggs kept at room temperature had greater development of their air chamber when compared to eggs kept under refrigeration; since these eggs responded positively to the test from the 15<sup>th</sup> day of storage, and the eggs stored in a refrigerated environment floated only after 30 days of storage (Table 3).

**Table 3.** Float test of quail eggs during the period of 30 days of storage at room (26.8°C) and refrigeration (10.9°C) temperatures.

	Temperatures							
Periods	Room	Refrigerated						
0	-	-						
5	-	-						
10	-	-						
15	+	-						
20	+	-						
25	+	-						
30	+	+						
	- = did not float; + = floated							

Water loss occurring soon after egg laying, as a result of the exchange of gases with the external environment, expands the air chamber, which reduces egg specific gravity (Poletti & Vieira, 2021), leading it to float. Similar to our results, Saccomani et al. (2019), when evaluating the effect of storage period and temperature on egg quality indicators, reported that with increasing storage time, eggs stored at room

temperature increase their floatage.

# Conclusion

Under experimental conditions, temperature and storage period are directly related to the internal quality of Japanese quail eggs. When stored at room temperature, quail eggs show decreases in internal quality during storage for up to 30 days. For the quail egg to reach the consumer market with better internal quality, it is recommended to store it under refrigeration.

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