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Carnauba wax coating preserves the internal quality of commercial eggs during storage

Revestimento com cera de carnaúba preserva a qualidade interna de ovos comerciais durante o armazenamento

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Highlights

We studied the effects of carnauba wax coating on quality and lipid oxidation of eggs.
Carnauba wax coating is able to maintain the internal quality of eggs during storage.
The effects of carnauba wax coating were most evident in eggs kept at 25°C.
Carnauba wax coating did not minimize the oxidative processes in the egg yolk.

Abstract

The objective of this study was to evaluate the internal quality and lipid oxidation of eggs coated with a carnauba wax-based product at different concentrations and stored for up to 28 days under two temperatures. For analysis of internal quality, the eggs were assigned to a completely randomized 3 x 4 factorial design (uncoated eggs (control); eggs coated with carnauba wax at 12% concentration (Aruá®); eggs coated with carnauba wax at 15% concentration (Aruá®); four storage periods - 7, 14, 21, and 28 days). Fifteen eggs from each treatment were evaluated in each storage period, with each egg representing one replicate, i.e., 300 eggs per storage temperature (10 and 25°C). Egg weight loss, yolk percent (%), albumen percent (%), Haugh unit, yolk index, and specific gravity were calculated. Lipid oxidation of the egg yolk was

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measured by thiobarbituric acid reactive substances (TBARS), using 10 eggs at time 0 (fresh) and 30 eggs in each storage period (7, 14, 21, and 28 days), in triplicate, under only one storage temperature (25°C). A total of five pools, consisting of two eggs each, were used for each treatment. Each pool was considered a replicate, and each treatment consisted of five replicates. The weight loss of the eggs stored at 10°C and 25°C during the storage period was, on average, 46.1% and 37.3% lower for the eggs coated with carnauba wax than in uncoated eggs, respectively. Overall, coated eggs, regardless the concentration of the wax (12 or 15%) had higher Haugh units, specific gravity, and yolk index than uncoated eggs, in both temperatures (10 and 25°C). Uncoated and coated eggs showed similar lipid oxidation values regardless of the storage period. On the other hand, eggs coated with solutions containing 15% wax showed less oxidation than eggs coated with 12% wax. The coating of commercial eggs with carnauba wax, both at concentrations of 12 and 15%, was effective in maintaining their internal quality during storage at both storage temperatures (10 and 25°C). Eggs stored at 25°C had lower quality traits during storage compared with eggs kept under refrigeration. Coating eggs with wax did not minimize the oxidative processes in the egg yolk.
Key words: Edible coating. Haugh unit. TBARS. Wax.

Resumo

O objetivo deste estudo foi avaliar a qualidade interna e a oxidação lipídica de ovos revestidos com um produto a base de cera de carnaúba, com diferentes concentrações, e estocados por até 28 dias sob condições de duas temperaturas. Para as análises de qualidade interna, os ovos foram distribuídos em um delineamento inteiramente casualizado em esquema fatorial 3 x 4 (ovos não revestidos (controle); ovos revestidos com cera de carnaúba a 12% (Aruá®); ovos revestidos com cera de carnaúba a 15% (Arua®); quatro períodos de estocagem - 7, 14, 21 e 28 dias). Quinze ovos de cada tratamento foram avaliados em cada período de estocagem, sendo considerado cada ovo uma repetição, totalizando 300 ovos por temperatura de estocagem (10 e 25°C). Para cada período foram calculados a perda de peso dos ovos, porcentagem de gema, porcentagem de albúmen, unidade Haugh, índice de gema e gravidade específica. A oxidação lipídica da gema dos ovos foi mensurada através das substâncias reativas ao ácido tiobarbitúrico (TBARS), utilizando 10 ovos para o tempo 0 (frescos) e 30 ovos em cada período de estocagem (7, 14, 21 e 28 dias), em triplicata, sob a temperatura de estocagem de 25°C. Um total de cinco pools, consistindo de dois ovos cada, foram utilizados para cada tratamento. Cada pool foi considerado uma repetição possuindo cada tratamento cinco repetições. A perda de peso dos ovos estocados a 10°C e a 25°C durante o armazenamento foi, em média, 46,1% e 37,3% mais baixo para os ovos revestidos com a cera de carnaúba em comparação aos ovos não revestidos, respectivamente. De maneira geral, os ovos revestidos, independentemente da concentração da cera (12 ou 15%) apresentaram maior unidade Haugh, gravidade específica e índice de gema comparado aos ovos não revestidos, em ambas as temperaturas (10 e 25°C). Os ovos não revestidos e revestidos apresentaram valores de oxidação lipídica similares independentemente do período de estocagem. Por outro lado, ovos revestidos com soluções contendo 15% de cera demonstraram menor oxidação do que os ovos revestidos com cera a 12%. O revestimento de ovos comerciais com cera de carnaúba, em ambas as concentrações de 12 e 15%, foi efetivo em manter a qualidade interna dos ovos durante o armazenamento em ambas as temperaturas (10 e 25°C). Ovos estocados a 25°C apresentaram menor qualidade comparado aos ovos mantidos sob refrigeração. O revestimento dos ovos com a cera não minimizou os processos oxidativos na gema do ovo.
Palavras-chave: Cera. Revestimento comestível. TBARS. Unidade Haugh.

Introduction

The growing concern of consumers about a nutritionally balanced and healthy diet has increased the consumption of egg protein in recent years. Eggs are a source of high-quality protein that contains nine essential amino acids and elements such as iron, phosphorus, vitamins, and essential fatty acids (Caner & Yüceer, 2015; Figueiredo et al., 2014). Moreover, the ratio of essential to total amino acids in eggs is higher than 40% and, within the recommended levels assigned by the Food and Agriculture Organization and World Health Organization (Sun, Liu, Yang, & Xu, 2019).

However, eggs are perishable and start deteriorating immediately after being laid. Losses in egg quality involve changes in chemical, physical, biological, and functional parameters (Hidalgo, Lucisano, Comelli, & Pompei, 1996), which impair not only the acceptability of the product by the consumer, but also its use by the food industry. Internal egg quality may particularly deteriorate under inappropriate storage conditions such as inadequate temperature, humidity, presence of CO2, and long storage periods (Chung & Lee, 2014; Samli, Agma, & Senkoylu, 2005).

Eggs naturally have a protective coating composed of glycoproteins and minor components such as hydroxyapatite crystals, polysaccharides, and lipids (Wellman-Labadie, Picman, & Hincke, 2008), reducing bacterial contamination and water loss through the eggshell. Many proteins present in the eggshell cuticle, including lysozyme C, ovotransferrin, ovocalyxin-32, cystatin, and ovoinhibitor, have antimicrobial activity (Rose-Martel, Du, & Hincke, 2012). Therefore, the cuticle acts as both a physical and chemical barrier by restricting water loss through the eggshell and preventing the penetration of undesirable microorganisms, respectively. Both processes are essential for preserving egg quality.

However, the legislations of several countries, including the USA, Australia, Japan (Hutchison et al., 2004), and Brazil (MAPA, 1990), determine that commercial eggs must be washed and sanitized before being sold. Although these procedures can reduce the microbial load on the eggshell, they may also damage the cuticle (Liu, Chen, Wu, Lee, & Tan, 2016). As a result, the eggs become susceptible to unfavorable conditions in terms of internal quality and bacterial penetration (Gole et al., 2014; Wardy, Torrico, No, Prinyawiwatkul, & Saalia 2010). Bacterial contamination is approximately 40% higher in eggs with damaged cuticles than in eggs with intact cuticles (Wilson, 2017).

The search for natural coatings that can seal the eggshell pores and mitigate deteriorative processes has increased due to the growing concern towards preserving egg quality during storage. These coatings are developed from polysaccharides, proteins, lipids, and combinations of thereof (Falguera, Quintero, Jiménez, Muñoz, & Ibarz, 2011). Among the products available are mineral oils, soy protein, whey protein, gluten, chitosan, corn-zein protein, and cellulose-based products, which are effective in reducing water loss and microorganism penetration through the egg pores (Almeida, Schneider, Yuri, Machado, & Gewehr, 2015; Molavi, Behfar, Ali Shariati, Kaviani, & Atarod, 2015; Yuceer & Caner, 2014).

Carnauba wax, which is extracted from the leaves of a Brazilian tropical palm tree, *Copernicia cerifera,* has been extensively

used in its refined form for coating fruits and vegetables and is considered safe for human consumption (Dou, 2004). The coating of fruits and vegetables with carnauba wax, in combination with additives such as cassava starch and glycerol, has shown satisfactory results in maintaining the quality and increasing the shelf life of products (Bhattacharjee & Dhua, 2018; Chiumarelli & Hubinger, 2014; Yang, Li, & Lu, 2018). Despite the evidence, no study to date has evaluated the use of carnauba wax to coat commercial eggs.

Based on this information, we hypothesized that coating eggs with carnauba wax can maintain their internal quality and stabilize lipid oxidation during storage. Therefore, the objective of this study was to evaluate the internal quality and lipid oxidation of eggs coated with a carnauba wax-based product at different concentrations and stored for up to 28 days under two temperatures.

Material and Methods

Location and experimental design

The experiment was carried out at the Federal University of Grande Dourados, Dourados, MS, Brazil. A total of 600 and 130 unfertilized white eggs from 35-week-old Bovans White hens were obtained from a commercial farm and used to perform internal quality and lipid oxidation analyses, respectively. To avoid egg cuticle damage which would favor the bacterial contamination, interfering in the evaluation of the effect of carnauba coating, the eggs were collected immediately after being laid and transported to the laboratory. Before the analyses initiated the eggs were screened for defects (crack, breakage, and surface cleanliness).

For analysis of internal quality, the unwashed eggs were weighed, labeled, and assigned to a completely randomized 3 x 4 factorial design (uncoated eggs (control); eggs coated with carnauba wax at 12% concentration (Aruá®); eggs coated with carnauba wax at 15% concentration (Aruá®); four storage periods - 7, 14, 21, and 28 days). Fifteen eggs from each treatment were evaluated in each storage period (7, 14, 21, and 28 days), with each egg representing one replicate, i.e., 300 eggs per storage temperature (10 and 25°C). Egg weight loss, yolk percent (%), albumen percent (%), Haugh unit, yolk index, and specific gravity were calculated.

Lipid oxidation of the egg yolk was measured by thiobarbituric acid reactive substances (TBARS), using 10 eggs at time 0 (fresh) and 30 eggs in each storage period (7, 14, 21, and 28 days), in triplicate, under only one storage temperature (25°C). A total of five pools, consisting of two eggs each, were used for each treatment. Each pool was considered a replicate, and each treatment consisted of five replicates.

Application of edible coating and storage

Coatings were composed of a commercial carnauba wax with vegetable resin-based solutions (colophony) (Aruá® Comércio e Serviço LTDA, São Paulo, Brazil) at concentrations of 12 and 15% (% of soluble components). Eggs were placed on rollers and the solutions were sprayed from a height of 15 cm above the eggs over the entire surface using a hand sprayer. Care was taken to ensure that each egg had a uniform coating with no visible defects. The control treatment did not receive carnauba wax treatment (uncoated eggs). The

eggs were air-dried at ambient conditions for approximately 30 min, and were then packed in pulp egg cartons. Cartons were then placed in two different Biochemical Oxygen Demand (BOD)-type climatic chambers according to storage temperature (10 and 25°C), under the same conditions of relative humidity (70%), for 28 days.

Egg quality

The eggs were weighed on day zero and at the end of each storage period, and egg weight loss (%) was calculated by the difference. After weighing, the eggs were analyzed for specific gravity by flotation in saline solutions of different concentrations (1.060, 1.065, 1.070, 1.075, 1.080, 1.085, and 1.090). The saline solutions were adjusted and calibrated periodically with the aid of a hydrometer.

After the evaluation of specific gravity, the eggs were broken on a flat surface and albumen height as well as yolk height and diameter were determined using a digital caliper. All measurements were taken in the central part of the egg. The Haugh unit was calculated based on the height of the albumen and egg weight, as described by Brant and Shrader (1958). The yolk index was obtained by the relation between its height and diameter.

Subsequently, yolk and eggshell were separated and individually weighed. Albumen weight was calculated by subtracting yolk weight and eggshell weight from egg weight. Yolk percentage and albumen percentage were calculated relative to the total egg weight.

Lipid oxidation in egg yolk

Determination of TBARS, expressed in milligrams of malondialdehyde per kilogram of yolk (mgMAD.kg-1), was performed according to the methodology adapted from Vyncke (1970). Two grams of the samples were homogenized with 8 mL of trichloroacetic acid at 15%. Subsequently, the tubes were placed in a homogenizer for 10 minutes and then centrifuged for 20 minutes at 3,000 rpm. Two mL of the obtained supernatant were homogenized with 2 mL of TBA solution (1% TBA, 15% TCA, and HCl 562.5 mM). The samples were then incubated at 100ºC for 15 minutes and, after cooling to room temperature, absorbance was measured in a spectrophotometer at 532 nm.

Statistical analyses

The effects of coating type, storage period, and their interaction were evaluated by analysis of variance. Means were compared by Tukey's test ($P < 0.05$) using SAS® University Edition (2017) (SAS Inst. Inc., Cary, NC, USA).

Results and Discussion

Egg quality

There was a significant interaction ($P < 0.05$) between coating type and storage period on egg weight loss and specific gravity in eggs stored at 10°C (Table 1). Overall, the sliced data show that although weight loss increased gradually during storage for all treatments, the weight loss in eggs coated with carnauba wax was, on average, 46.1% lower ($P < 0.05$) than

that in uncoated eggs. Uncoated eggs had a lower specific gravity after 21 days of storage ($P < 0.05$), whereas coated eggs differed only in the last storage period (28 days). Within each storage period, coated eggs, regardless of the concentration, showed a higher specific gravity than uncoated eggs after 7 days of storage (Table 2). There was a significant effect ($P < 0.05$) of coating type and storage period on Haugh unit and yolk index. Regardless of the storage period, eggs coated with carnauba wax had higher Haugh unit and yolk index values than uncoated eggs. Moreover, the eggs had lower Haugh unit values after 21 days of storage, regardless of the coating type. Yolk percentage and albumen percentage were affected ($P < 0.05$) by the storage period, in which albumen % decreased and yolk % increased with increasing storage period (Table 1).

There was a significant interaction ($P < 0.05$) between coating type and storage period on all variables associated with internal quality (weight loss, Haugh Unit, Yolk Index, albumen %, yolk %, and specific gravity) in eggs stored at 25°C (Table 3). The sliced data show that, within each storage period, coating with carnauba wax decreased ($P < 0.05$) weight loss by 69.4, 60.6, 67.5, and 58.8%, on average, in eggs stored for 7, 14, 21, and 28 days, respectively. Furthermore, coating with solutions containing wax at 15% was more efficient in reducing egg weight loss up to 21 days, with results similar to those coated with 12% wax at 28 days. Coated eggs had higher Haugh units at 14 and 21 days and higher yolk index values at 7, 14, and 21 days ($P < 0.05$) than uncoated eggs. At 28 days, eggs coated with carnauba wax at 15% concentration showed a response similar to that of uncoated eggs for both variables. Coated eggs had higher albumen % values at 21 and 28 days of storage, but lower yolk % at 28 days of storage. They also showed a higher specific gravity than uncoated eggs before 21 days of storage, whereas after 28 days, the eggs showed a similar response. There was a gradual increase in egg weight loss (%) and yolk % with increasing storage period, regardless of the treatment (coated or uncoated). On the other hand, the remaining variables (Haugh units, Yolk Index, albumen %, and specific gravity) decreased progressively with increasing storage period (Table 4).

Keeping fresh eggs under refrigeration is a feasible strategy to increase their shelf life, even though this is not possible in many regions and countries. Therefore, the use of edible coating is viable alternative to reduce egg weight loss during storage and to maintain internal egg quality. Different types of coatings, such as mineral oil, beeswax, milk and soy proteins, and chitosan, have proved to be effective in maintaining the quality of eggs during storage (Sharaf Eddin, Ibrahim, & Tahergorabi, 2019).

Table 1
Internal egg quality of commercial laying hens coated with different concentrations of carnauba wax and stored at 10°C, for 28 days

Treatments	Weight loss (%)	Haugh unit	Yolk index	% of albumen	% of yolk	Specific gravity ($g.cm^{-3}$)
Edible coating						
0 (control)	2.55	80.69b	0.377b	61.59	28.78	1.062
12% wax	1.38	84.50a	0.393a	62.31	28.09	1.074
15% wax	0.97	84.31a	0.401a	62.05	28.39	1.074
Storage Period						
7 days	0.60	84.77a	0.396ab	63.45a	27.29c	1.077
14 days	1.38	83.63a	0.383bc	62.33ab	28.10bc	1.074
21 days	1.52	86.52a	0.408a	61.53bc	28.72ab	1.071
28 days	3.04	77.75b	0.373c	60.63c	29.58a	1.058
SEM[1]	0.019	0.433	0.001	0.141	0.124	0.001
			ANOVA			
Coating (C)	<0.0001	<0.0001	<0.0001	0.3354	0.2853	<0.0001
Period (P)	<0.0001	0.0233	<0.0001	<0.0001	0.0003	<0.0001
Interaction C x P	<0.0001	0.1912	0.2637	0.6885	0.8220	0.0427

Means with different letters within a column are significantly different (P <0.05) after Tukey test.
[1]SEM = standard error of the mean.

Table 2
Deployment of interaction between coating and storage period on weight loss and specific gravity in eggs from commercial laying hens coated with different concentrations of carnauba wax and stored at 10°C, for 28 days

	Storage Period (days)			
	7	14	21	28
Edible coating		Weight loss (%)		
0 (control)	1.06dA	2.03cA	2.65bA	4.47aA
12% wax	0.44dB	1.42bB	1.02cB	2.65aB
15% wax	0.32cC	0.70bC	0.88bB	2.00aC
		Specific gravity ($g.cm^{-3}$)		
0 (control)	1.071aB	1.067aB	1.061bB	1.051cB
12% wax	1.080aA	1.078aA	1.077aA	1.060bA
15% wax	1.080aA	1.079aA	1.075aA	1.062bA

Means with different lowercase letters within a line are significantly different after Tukey test (P <0.05).
Means with different upper letters within a column are significantly different after Tukey test (P <0.05).

Table 3
Internal egg quality of commercial laying hens coated with different concentrations of carnauba wax and stored at 25°C, for 28 days

Treatments	Weight loss (%)	Haugh unit	Yolk index	% of albumen	% of yolk	Specific gravity ($g.cm^{-3}$)
Edible coating						
0 (control)	6.87	59.37	0.262	58.30	31.44	1.053
12% wax	2.88	67.99	0.314	61.08	29.23	1.060
15% wax	2.25	68.26	0.317	60.40	29.96	1.065
Storage Period						
7 days	1.41	79.06	0.365	62.47	28.16	1.069
14 days	3.10	72.28	0.312	60.07	29.95	1.061
21 days	4.45	59.52	0.289	59.20	30.88	1.056
28 days	7.03	49.27	0.226	57.97	31.84	1.051
SEM1	0.055	0.398	0.002	0.137	0.134	0.001
ANOVA						
Coating (C)	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Period (P)	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Interaction C x P	<0.0001	0.0035	<0.0001	<0.0001	0.0002	<0.0001

[1]SEM = standard error of the mean.

Carnauba wax, which is extracted from the leaves of a Brazilian tropical palm tree, *Copernicia cerifera,* has been extensively used around the world for coating fruits and vegetables (Singh et al., 2019; Yang et al., 2018). The incorporation of carnauba wax into foods and its use as an additive and coating material is considered safe by several food safety organizations (Freitas et al., 2019). It is believed that the leaves of this palm are covered by a waxy layer as an adaptation to dry environments. This coating forms a semipermeable barrier to oxygen, carbon dioxide, and water, thereby reducing respiration rates, oxidation reactions, and water loss, thus contributing to food preservation (Falguera et al., 2011). Moreover, coating with carnauba wax enhances the gloss and appearance of foods, increasing customer acceptance.

Carnauba wax was an efficient physical barrier by sealing the pores and reducing the loss of components through the eggshell during storage, confirming our hypothesis about the maintenance of internal egg quality. In our study, the effects were most evident in eggs kept at 25°C. Many chemical reactions between the internal components of an egg occur soon after laying and during storage, with consequent quality loss. These reactions include the dissociation of carbonic acid, which is a component of the buffer system in

Table 4
Deployment of interaction between coating and storage period on weight loss, haugh unit, yolk index, albumen, yolk and specific gravity in eggs of commercial laying hens coated with different concentrations of carnauba wax and stored at 10°C, for 28 days

Edible coating	Storage Period (days)			
	7	14	21	28
		Weight loss (%)		
0 (control)	2.63dA	5.20cA	8.09bA	11.56aA
12% wax	0.91cB	2.63bB	3.11bB	4.86aB
15% wax	0.70dC	1.47cC	2.15bC	4.67aB
		Haugh unit		
0 (control)	75.42aA	63.86bB	52.25cB	45.95dB
12% wax	81.29aA	73.94bA	61.29cA	56.54dA
15% wax	80.47aA	79.03aA	65.03bA	47.41cB
		Yolk index		
0 (control)	0.341aB	0.280bC	0.239cC	0.189dB
12% wax	0.377aA	0.315bB	0.293bcB	0.272cA
15% wax	0.376aA	0.340bA	0.335bA	0.218cB
		Albumen (%)		
0 (control)	61.88aB	60.57aA	56.71bB	54.05cB
12% wax	63.94aA	59.40bA	60.58bA	60.42bA
15% wax	61.59aB	60.24aA	60.32aA	59.46aA
		Yolk (%)		
0 (control)	28.55cA	29.51cA	32.50bA	35.18aA
12% wax	26.72bB	30.47aA	29.93aA	29.82aB
15% wax	29.20aA	29.90aA	30.22aA	30.52aB
		Specific gravity ($g.cm^{-3}$)		
0 (control)	1.061aB	1.050bC	1.050bC	1.050bA
12% wax	1.071aA	1.061bB	1.054cB	1.052cA
15% wax	1.073aA	1.071aA	1.063bA	1.052cA

Means with different lowercase letters within a line are significantly different after Tukey test (P <0.05).
Means with different upper letters within a column are significantly different after Tukey test (P <0.05).

the albumen, into water and carbon dioxide. As a result, both molecules are transferred to the environment through the pores of the eggshell (Brake et al., 1997). Therefore, the rate of weight loss during storage is an important method to analyze egg quality.

The loss of the above-mentioned elements has undesirable consequences, including increased pH and reduced albumen viscosity, with consequent losses in egg weight (Caner & Cansiz, 2008). The reduced albumen viscosity is associated with changes in its protein structure, including increased levels of clusterin and ovoinhibitor and a disorder of the ovalbumin structure (Sheng, Huang, Wang, Xu, Hammad, & Ma, 2018). The extent of these changes is quantified by the Haugh unit, which associates albumen height with egg weight. Moreover, the continuous loss of moisture and carbon dioxide to the environment progressively increases the size of the air chamber, which reduces the specific gravity of eggs (Xu, Zhang, Lv, Chi, Wu, & Shao, 2017). Coating eggs with carnauba wax resulted in less weight loss and, consequently, fewer variations in albumen structure, which led to higher Haugh unit values as well as higher values of specific gravity.

The breakdown of structural proteins in the vitelline membrane, coupled with differences in osmotic pressure, favors the movement of water from the albumen to the yolk, which reduces the integrity of the membrane (Akter, Kasim, Omar, & Sazili, 2014). This fact is compatible with the reduction of albumen % and the increase of yolk % during storage. Coating slows the rise in osmotic pressure by reducing the structural changes in the albumen, ensuring improved yolk quality during storage (Xu, Wang, Ren, & Wu, 2018).

Eggs kept at room temperature showed lower internal quality during storage. The reduction of internal quality was lower in eggs kept under refrigeration, since lower temperatures minimize the occurrence of degradation reactions. Feddern, Prá, Mores, Nicoloso, Coldebella and Abreu (2017) observed that eggs stored for 9 weeks under refrigeration (0-5°C) had a quality similar to those kept at room temperature (20-35°C) for only 3 weeks, which demonstrates the influence of temperature on quality parameters of eggs. In addition to the interference with the quality parameters, higher storage temperatures can increase the penetration of bacteria into eggs (Wang & Slavik, 1998). In this context, Menezes, Lima, Medeiros, Oliveira and Evêncio (2012) distinguished temperature and humidity as the two main factors affecting the egg quality during storage. The benefits of carnauba wax coating on egg quality parameters were evident in eggs kept at 25°C. Therefore, carnauba wax demonstrated its potential for minimizing the main factors responsible for accelerating quality loss.

Besides favoring physical and chemical changes of both albumen and yolk, environmental and storage conditions can increase the internal contamination with undesirable microorganisms, which accelerates the degradation processes (Xu et al., 2017). Thus, coating with carnauba wax has proven to be an efficient barrier against microorganisms, maintaining a high internal egg quality.

Lipid oxidation in egg yolk

There was no significant interaction (P > 0.05) between the factors on lipid oxidation (TBARS) of the egg yolk. However, the type of coating affected (P < 0.05) malondialdehyde concentrations. Uncoated and coated eggs showed similar lipid oxidation values regardless of the storage period. On the other hand, eggs coated with solutions containing 15% wax showed less oxidation than eggs coated with 12% wax (Table 5).

Table 5
Effect of carnauba wax coating on thiobarbituric acid reactive substances (TBARS[1]) values on egg yolk of laying hens stored for 28 days

Storage period (days)							
Edible coating	0	7	14	21	28	Average	SEM
0 (control)	0.067	0.139	0.120	0.128	0.082	0.113ab	0.013
12% wax	0.067	0.182	0.117	0.174	0.088	0.145a	0.015
15% wax	0.067	0.074	0.111	0.096	0.109	0.098b	0.010
Average	0.067	0.132	0.116	0.133	0.093		0.008
SEM[2]	0.006	0.018	0.011	0.019	0.001	0.008	
ANOVA							
Coating (C)	0.046						
Days (D)	0.082						
Interaction C x D	0.235						

[1]Expressed in milligrams of malondialdehyde per kilogram of yolk (mgMAD.kg^{-1}).
[2]SEM = standard error of the mean.
Means with different letters within a column are significantly different after Tukey test (P <0.05).

In addition to influencing the internal quality parameters of eggs, the coatings may be able to minimize the oxidation process of lipids in the yolk. Studies have shown that the control of gas exchange between the fruit and the environment results in less oxygen available for respiration and, thus, for metabolic processes (Bonilla, Atarés, Vargas, & Chiralt, 2012), which delays lipid oxidation. Despite the reduction of weight loss in coated eggs, which demonstrated that carnauba wax was effective in sealing the pores of the eggshell and controlling the gas exchange between the internal and the external environment, it was not possible to observe a reduction in lipid oxidation in the yolks of coated eggs. This finding does not support our hypothesis about the influence of carnauba wax on lipid stability. The antioxidant capacity of carnauba wax is still controversial in the literature. Although it is evidenced that carnauba wax coating is a potential physical barrier, some studies have shown that extracts from wax powder have antioxidant and antifungal activities (Andrade et al., 2018). However, Zhang, Simpson and Dumont (2018) observed no change in

antioxidant activity when using carnauba wax as an additive in gelatin films.

In a study on the use of mixed coatings containing carnauba wax to preserve fresh-cut apples, Chiumarelli and Hubinger (2014) reported increased respiration rates with increasing inclusion levels (%) of wax. This result indicated that the concentration of this component might facilitate gas exchange with the environment. Despite these findings and considering that the coating in this experiment contained only carnauba wax, eggs coated with 15% carnauba wax had a lower amount of malondialdehyde compared with eggs coated with wax at 12%, which were in turn similar to uncoated eggs.

Conclusion

The coating of commercial eggs with carnauba wax, both at concentrations of 12 and 15%, was effective in maintaining their internal quality during storage at both storage temperatures (10 and 25°C). Eggs stored at 25°C had lower quality traits during storage compared with eggs kept under refrigeration. Coating eggs with wax did not minimize the oxidative processes in the egg yolk.

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Conflict of interest declaration

The authors declared no conflict of interest.

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