















Effectiveness of an andiroba oil bioproduct in maintaining egg quality at room temperature

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Abstract

The aim of this study was to evaluate the effectiveness of a bioproduct based on andiroba oil (AO; *Carapa guianensis*) in preserving the physical, bacteriological, and chemical qualities of chicken eggs stored at room temperature for 7 days. Eggs were treated with different concentrations of AO (1–15%), two application methods (spray or immersion), and two exposure times (15 and 30 s). The bioproduct effectively reduced microbial load, particularly *Escherichia coli*, *Staphylococcus aureus*, and *Salmonella spp.*, and improved the conservation of egg weight, albumen and yolk height, shell thickness, and Haugh unit (HU). Higher oil concentrations, especially at 15% with immersion for 30 s, provided the best results by forming a protective film that minimized gas and moisture exchange. Additionally, the coating reduced lipid oxidation and maintained yolk lipid content, confirming the protective and antioxidant action of the bioproduct. Although higher oil concentrations slightly affected sensory acceptance, the product proved to be a promising natural alternative to refrigeration for short-term egg storage in tropical conditions.

Keywords: Amazon; andiroba oil; antimicrobial; *Carapa guianensis*; coating; egg.

Practical Application: Natural coating with andiroba oil (AO) preserves the egg quality without refrigeration.

1 INTRODUCTION

Eggs are among the most complete foods in the human diet due to their nutritional composition rich in vitamins, minerals, fatty acids, and proteins (Eddin et al., 2019; Kusum et al., 2018). However, their quality can deteriorate during storage depending on time, temperature, humidity, air circulation, and packaging type (Kusum et al., 2018; Pires et al., 2020). Time is the main factor affecting these conditions, as prolonged storage, especially at room temperature, accelerates the loss of internal quality. Refrigeration effectively delays these changes, maintaining freshness, reducing microbial risk, and extending shelf life (Rêgo et al., 2012; Salgado et al., 2018).

In many countries, such as Brazil, the United Kingdom, Sweden, and Spain, legislation does not require egg refrigeration, only recommending it to extend shelf life (Berkhoff et al., 2020; Pires et al., 2020). In these regions, eggs are often kept at room temperature from production to consumption, mainly to reduce storage costs (Lana et al., 2017). Nevertheless, such practices demand rapid distribution and commercialization to prevent deterioration and maintain food safety (Rêgo et al., 2012).

To address this issue, several studies have explored techniques and bioproducts to preserve eggs without refrigeration. Surface coatings have proven particularly promising, acting as artificial cuticles that reduce gas exchange and maintain internal and external quality for longer periods (Biladeau & Keener, 2009; Brasil et al., 2019; Pissinati et al., 2014). Oil-based coatings stand out for their hydrophobicity and sealing capacity, minimizing weight loss and preserving internal characteristics during storage (Brasil et al., 2019; Waimaleongora-Ek et al., 2009).

The Brazilian Amazon offers a wide diversity of native species with potential for developing such bioproducts (Clement et al., 2005). Among them, AO (*Carapa guianensis* Aubl.) presents remarkable physicochemical properties, antibacterial, antioxidant, antiparasitic, antiseptic, antiviral, and emollient (Carvalho et al., 2019; Meccia et al., 2013). Its composition includes fatty acids, triterpenes, tannins, and alkaloids that confer broad biological activity and commercial value (Sousa et al., 2022). Therefore, the aim of this study was to evaluate the efficiency of a bioproduct based on andiroba oil (BBAO) in preserving the quality of eggs stored at room temperature for 7 days.

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1.1 Relevance of the work

This study presents an innovative and sustainable approach to egg preservation using a natural bioproduct formulated with AO, a native Amazonian resource with antimicrobial and antioxidant properties. Its application provides an alternative to refrigeration, maintaining egg quality under tropical conditions while adding socioeconomic value to regional biodiversity. The findings support the potential of AO bioproducts for food preservation, contributing to environmental sustainability, food security, and the valorization of Amazonian bioresources.

2 MATERIALS AND METHODS

The current experiment was conducted at the Faculty of Agrarian Sciences of the Universidade Federal do Amazonas, located in Manaus (AM), Brazil. All experimental procedures were performed in accordance with the Local Experimental Animal and Human Care Committee and were approved by the institutional ethics committee.

2.1 Andiroba oil acquisition and bioproduct based on andiroba oil production

The AO was prepared by family producers in the municipality of Manaus, Amazonas, Brazil. The oil was extracted using a traditional method without solvents (Sousa et al., 2019). The seeds were collected, cooked until softened, cooled to room temperature, separated from the shell and endosperm, and manually pressed two to three times daily, yielding approximately 500 g of oil resin per kg of seed. For this step, the endosperm was placed in a stainless-steel container on an inclined plane to drain the oily fraction, and finally, crude AO was obtained. Subsequently, crude AO was strained to obtain the pure AO, which was stored in hermetically sealed containers.

A sample fraction of the pure oil (approximately 500 mL) was sent to the laboratory for physical-chemical determinations and chemical characterization of the oil substances according to the methods described by AOCS (2022) and Sousa et al. (2022). The physicochemical analysis of the AO revealed averages of $2.37 \pm 0.28\%$ free fatty acid content, acidity value of 3.26 ± 0.55 mg NaOH/g, peroxide value of 1.53 ± 0.32 meq/1,000 g, saponification value of 186.92 ± 0.24 mg KOH/g, iodine value of 87.68 ± 0.53 g I₂/100 g, pH of 4.63 ± 0.02 (mg/100 g), and moisture value of $0.001 \pm 0.11\%$. The AO composition analysis identified eight chemical constituents, accounting for 99.98% of the oil. Saturated fatty acids comprised 42.47%, monounsaturated fatty acids made up 45.43%, polyunsaturated fatty acids constituted 8.65%, and triterpenoids accounted for 3.43%. The primary fatty acids identified in AO were oleic (45.43%), palmitic (28.44%), stearic (10.05%), and linoleic acids (8.65%).

The BBAO was prepared by initially placing the AO in a 1-L graduated plastic container, following the predetermined concentration for each treatment. While stirring, a nonionic surfactant (Tween 20) was gradually added to the AO, after which distilled water was slowly incorporated under continuous agitation to ensure complete homogenization of the mixture. The resulting BBAO solutions were then transferred to appropriate

containers depending on the intended application method—either 1-L plastic bottles or 500-mL spray bottles. The formulation of each BBAO sample followed Equation 1:

$$\text{BBAO} = \text{AO} + 10\text{NIS} + \text{DW} \quad (1)$$

Where:

AO: the percentage of AO according to the treatment;

10NIS: a fixed 10% of nonionic surfactant (Tween 20);

DW: the percentage of distilled water required to complete 100%.

2.2 Egg sources, experimental design, and BBAO application

Eggs were sourced from Hissex Brown hens (56 weeks of age) housed in cages (1.00 × 0.45 × 0.45 m) with a stocking density of 0.16 birds/m². The hens were managed using standard diets formulated based on the recommendations of Rostagno et al. (2017). They had access to water *ad libitum* and were managed according to the standard guidelines provided in the strain manual. The eggs were arranged in a completely randomized design in a factorial arrangement (2 + 4 × 2 × 2), where the treatments were constituted by a positive (fresh egg, without storage and freshly collected) and a negative (egg storage by 7 days without application of BBAO) control, four levels of AO in the bioproduct formula (1, 5, 10, and 15%), two application methods of the BBAO (spray or dip) and two exposure periods of the egg to the BBAO during the application (15 and 30 s), totaling 18 treatments, with 25 eggs each, where the egg was considered as a replicate. For the application of BBAO, the eggs were immersed in a sterilized container containing the BBAO or sprayed on all its surfaces using a sterilized manual sprayer containing the BBAO according to the pre-established treatments. Then, the eggs were stored for 7 days at room temperature with controlled environmental conditions presenting averages of 22.4°C (72.32°F) of temperature and 55% of air relative humidity.

2.3 Physical quality

Five eggs from each treatment underwent a physical quality analysis according to the methods described by Brasil et al. (2019). These included weighing the eggs with precision, measuring yolk and albumen heights and circumferences, determining yolk and albumen percentages relative to the egg weight, measuring the pH of yolk and albumen, assessing yolk color on a scale from 1 to 15, and evaluating eggshell, including percentage in relation to egg weight, and thickness in three regions (basal, meridional, and apical). HU was calculated using egg weight (W) and albumen height (H) values the Equation 2:

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

2.4 Bacteriological quality

The bacteriological analysis procedures were conducted according to the Brazilian standard normative (Brazil, 2018). Standardized samples were prepared by collecting the internal content of five eggs from each treatment, homogenizing it for 60 s, and creating dilutions of 10⁻¹ and 10⁻² with 1% buffered peptone water. For total mesophyll enumeration, selected dilutions were inoculated into Petri plates with

Plate Count Agar and incubated at 36°C for 48 h. The outcome was expressed as Colony Forming Units per 1.0 g of sample (CFU/g). To enumerate *Escherichia coli*, dilutions were inoculated into test tubes and incubated at 36°C. Positive outcomes were confirmed by subsequent testing and expressed as the most probable number per gram (MPN/g). *Staphylococcus aureus* count was determined by inoculating dilutions onto Baird-Parker Agar plates, followed by incubation at 36°C for 48 h. The outcome was expressed as Colony Forming Units per 1.0 g of sample (CFU/g). *Salmonella* spp. analysis was conducted through pre-enrichment, selective broths, and PCR testing for diagnosis and confirmation.

2.5 Chemical composition and lipid oxidation (TBARS)

Five eggs from each treatment underwent the chemical content analysis, which included assessing moisture, minerals, fats, and proteins. These analyses were conducted following the methods recommended by the Association of Official Analytical Chemists (AOAC, 2019). The other five eggs from each treatment were used to assess the TBARS levels, which indicate the degree of lipid oxidation. At the end of the 7-day storage period, the eggs were cracked, and their yolks were separated and frozen. Subsequently, the frozen yolks underwent freeze-drying, where water and other solvents were removed through sublimation, bypassing the liquid state. The dehydrated yolks were then analyzed for lipid oxidation (TBARS) using a modified version of the methodology described by Vyncke (1970) and adapted by Ramanathan and Das (1992).

2.6 Sensory analysis

Three eggs from each treatment, totaling 54 eggs, were used for sensory analysis. These eggs were hard-boiled, cooled, and prepared by removing the hot water and placing them in cool water for 3 min. The eggs were then peeled, cut into halves, further divided into quarters, and placed on plates with random identification numbers known only to the researchers (Hayat et al., 2010). Twenty untrained judges conducted the sensory analysis after a 3-h period of not consuming food or smoking. To prevent bias from eggshell colors, the judges did not have visual contact with the eggshells. They assessed the eggs for appearance, yolk color, aroma, and flavor using a continuous unstructured line intensity scale ranging from 0 to 9 (Berkhoff et al., 2020).

2.7 Statistical analyses

The data were initially assessed for normality, and necessary transformations were applied. Subsequently, a one-way ANOVA was conducted using R software (version 4.1.3) following Logan's (2010) guidelines. Tukey's honestly significant difference test was used to identify significant differences among mean values, and results are presented as means, with statistical significance defined at $p \leq .05$.

3 RESULTS AND DISCUSSION

3.1 Physical quality of the eggs

Tables 1 and 2 show the average results for the physical quality of eggs. The use of BBAO had a significant ($p \leq .05$) impact

on various analyzed variables, except for yolk and albumen pH, and yolk percentage. Higher levels of AO in BBAO resulted in significantly better conservation of egg weight, albumen percentage, yolk and albumen height, yolk color, eggshell thickness, and HU in eggs stored for 7 days compared to those also stored but without BBAO (the negative control). However, yolk and albumen diameters, as well as eggshell percentages, increased in eggs coated with BBAO, regardless of the technique and application time used. In summary, the application of BBAO, while not maintaining the same physical quality as fresh eggs, improved the conservation of these characteristics when eggs were stored at room temperature.

In general, higher levels of AO in the BBAO contributed to the development of an additional protective film on the eggshell, leading to better conservation of its physical attributes, just like the dip as the better application method and 30 s as the better time application. This protective cuticle covers the entire shell surface, safeguarding pore openings and hindering the passage of water

Table 1. Egg weight and percentages of yolk, albumen, and shell of eggs submitted to a BBAO with different levels of AO, and applied using different methods and times.

Factors	Variables ¹			
	EW	Y	A	S
AO level (LV)				
Positive control	60.12 ± 0.02 ^a	30.05 ± 0.02	55.74 ± 0.01 ^a	9.64 ± 0.04 ^c
Negative control	56.25 ± 0.12 ^b	32.13 ± 0.04	45.74 ± 0.01 ^c	11.17 ± 0.02 ^a
1	56.68 ± 0.11 ^c	30.93 ± 0.05	51.68 ± 0.11 ^b	10.61 ± 0.05 ^b
5	57.26 ± 0.05 ^c	31.37 ± 0.06	51.07 ± 0.04 ^b	11.09 ± 0.03 ^a
10	59.64 ± 0.03 ^b	30.58 ± 0.03	49.54 ± 0.03 ^b	10.39 ± 0.04 ^b
15	58.79 ± 0.04 ^b	29.88 ± 0.02	50.85 ± 0.05 ^b	10.35 ± 0.01 ^b
Technique (TQ)				
Positive control	60.12 ± 0.02 ^a	30.05 ± 0.02	55.74 ± 0.01 ^a	9.64 ± 0.04 ^c
Negative control	56.25 ± 0.12 ^c	32.13 ± 0.04	45.74 ± 0.01 ^c	11.17 ± 0.02 ^a
Spray	57.42 ± 0.05 ^b	31.04 ± 0.06	49.59 ± 0.05 ^b	10.20 ± 0.05 ^b
Dip	58.76 ± 0.03 ^b	30.34 ± 0.05	51.98 ± 0.04 ^b	11.02 ± 0.03 ^a
Time (TM)				
Positive control	60.12 ± 0.02 ^a	30.05 ± 0.02	55.74 ± 0.01 ^a	9.64 ± 0.04 ^c
Negative control	56.25 ± 0.12 ^c	32.13 ± 0.04	45.74 ± 0.01 ^c	11.17 ± 0.02 ^a
15	58.40 ± 0.03 ^b	30.83 ± 0.02	49.29 ± 0.06 ^b	10.85 ± 0.02 ^b
30	57.78 ± 0.02 ^b	30.55 ± 0.04	52.28 ± 0.05 ^b	10.37 ± 0.03 ^b
Effect				
LV ²	0.05	0.86	0.05	0.01
TQ ²	0.05	0.54	0.05	0.01
TM ²	0.05	0.80	0.03	0.01
LV*TQ ³	0.45	0.57	0.83	0.34
LV*TM ³	0.90	0.77	0.35	0.13
TQ*TM ³	0.69	0.19	0.56	0.22
LV*TQ*TM ³	0.75	0.72	0.74	0.62
CV, %	4.15	5.62	8.52	6.38

¹EW: Egg weight (g); Y: yolk (%); A: albumen (%); S: shell (%); ²means followed by different letters in the column differ by the Tukey test ($p \leq .05$); ³a $p \leq .05$ indicates a direct interaction between the evaluated factors; CV: coefficient of variation.

and bacteria, consequently conserving for more time the quality of the eggs (Eddin et al., 2019; Muñoz et al., 2015; Pires et al., 2020). The increased eggshell thickness in BBAO-treated eggs supports this observation, as do the results related to albumen percentage, yolk and albumen height, yolk color, and the lowest weight loss (or highest egg weight retention) in eggs stored for 7 days with BBAO. These findings are consistent with the reports from Mendonça et al. (2013) and Salgado et al. (2018), which also showed that coated eggs experienced less weight loss compared to uncoated eggs. Lipophilic coatings, as suggested by Biladeau and Keener (2009), prevent water ingress through the eggshell, aiding in the conservation of the internal egg content.

3.2 Bacteriological quality

As shown in Table 3, fresh eggs contained no *Escherichia coli*, *Staphylococcus aureus*, or mesophiles, whereas eggs stored for 7 days without BBAO exhibited significantly higher ($p \leq .05$) bacterial counts. BBAO effectively reduced or eliminated bacteria at room temperature, especially at higher AO levels. Eggs immersed in BBAO for 30 s showed the lowest bacterial concentrations, with the spray method also performing well.

Salmonella spp. was detected in eggs stored without treatment and in those coated by spray using concentrations of up to

10% AO, regardless of exposure time. However, eggs treated with 15% AO, whether by spray or immersion, and all eggs treated by immersion at any concentration showed no presence of *Salmonella spp.* These results demonstrate that immersion provides superior microbial protection compared to spraying and that increasing the concentration of AO enhances the antimicrobial effect, completely preventing *Salmonella* contamination under the highest concentration.

The decline in the physical quality observed in eggs without BBAO or with lower AO levels, particularly when applied by spraying and for shorter durations, likely results from insufficient surface coverage and greater exposure to microorganisms. Conversely, higher AO concentrations, especially when applied by dipping for longer times, reduced bacterial counts, indicating the biofilm's effectiveness as a protective barrier (Eddin et al., 2019; Pires et al., 2020). AO exhibits inhibitory activity against *Staphylococcus aureus* and *Escherichia coli* and may limit microbial adhesion (Conde et al., 2015; Meccia et al., 2013). Thus, BBAO acts as an efficient barrier, minimizing water loss and microbial penetration.

3.3 Chemical composition and lipid oxidation (TBARS)

The chemical composition of the eggs, as shown in Table 4, revealed that an increase in AO levels within BBAO resulted in a

Table 2. Physical quality of eggs submitted to a BBAO with different levels of AO, and applied using different methods and times.

Factors	Variables ¹								
	YH	AH	YD	AD	YC	YPh	APh	ST	HU
AO level (LV)									
Positive control	18.25 ± 0.12 ^a	7.50 ± 0.12 ^a	38.00 ± 0.13 ^c	70.75 ± 0.17 ^c	7.25 ± 0.13 ^a	7.11 ± 0.19	7.32 ± 0.15	0.37 ± 0.05 ^c	100.00 ± 0.00 ^a
Negative control	16.73 ± 0.11 ^c	5.38 ± 0.15 ^c	41.55 ± 0.17 ^b	108.14 ± 0.15 ^b	6.14 ± 0.17 ^c	7.06 ± 0.16	7.35 ± 0.14	0.38 ± 0.08 ^c	92.66 ± 0.13 ^c
1	17.12 ± 0.14 ^b	6.02 ± 0.17 ^b	42.93 ± 0.16 ^b	101.04 ± 0.16 ^b	6.62 ± 0.16 ^b	7.11 ± 0.18	7.24 ± 0.16	0.44 ± 0.08 ^b	95.93 ± 0.13 ^b
5	17.02 ± 0.15 ^b	5.93 ± 0.15 ^c	43.89 ± 0.13 ^a	110.50 ± 0.11 ^b	6.54 ± 0.13 ^b	7.13 ± 0.19	7.24 ± 0.16	0.44 ± 0.07 ^b	95.37 ± 0.15 ^b
10	17.04 ± 0.12 ^b	5.54 ± 0.17 ^c	43.77 ± 0.15 ^a	109.81 ± 0.14 ^b	6.66 ± 0.13 ^b	7.08 ± 0.19	7.18 ± 0.15	0.48 ± 0.05 ^a	93.27 ± 0.13 ^b
15	18.08 ± 0.11 ^a	5.47 ± 0.13 ^c	44.43 ± 0.14 ^a	115.14 ± 0.13 ^a	6.85 ± 0.14 ^b	7.15 ± 0.13	7.24 ± 0.18	0.47 ± 0.09 ^a	94.48 ± 0.11 ^b
Technique (TQ)									
Positive control	18.25 ± 0.12 ^a	7.50 ± 0.12 ^a	38.00 ± 0.13 ^c	70.75 ± 0.17 ^b	7.25 ± 0.13 ^a	7.11 ± 0.19	7.32 ± 0.15	0.37 ± 0.05 ^c	100.00 ± 0.00 ^a
Negative control	16.73 ± 0.11 ^c	5.38 ± 0.15 ^b	41.55 ± 0.17 ^b	108.14 ± 0.15 ^a	6.14 ± 0.17 ^c	7.06 ± 0.16	7.35 ± 0.14	0.38 ± 0.08 ^c	92.66 ± 0.13 ^c
Spray	17.52 ± 0.15 ^b	5.96 ± 0.13 ^b	43.16 ± 0.17 ^a	107.86 ± 0.13 ^a	6.76 ± 0.13 ^b	7.14 ± 0.15	7.30 ± 0.15	0.49 ± 0.02 ^a	95.36 ± 0.12 ^b
Dip	17.11 ± 0.16 ^b	5.66 ± 0.15 ^b	44.35 ± 0.12 ^a	110.38 ± 0.13 ^a	6.58 ± 0.18 ^b	7.09 ± 0.15	7.15 ± 0.11	0.42 ± 0.03 ^b	94.17 ± 0.12 ^b
Time (TM)									
Positive control	18.25 ± 0.12 ^a	7.50 ± 0.12 ^a	38.00 ± 0.13 ^c	70.75 ± 0.17 ^b	7.25 ± 0.13 ^a	7.11 ± 0.19	7.32 ± 0.15	0.37 ± 0.05 ^b	100.00 ± 0.00 ^a
Negative control	16.73 ± 0.11 ^c	5.38 ± 0.15 ^c	41.55 ± 0.17 ^b	108.14 ± 0.15 ^a	6.14 ± 0.17 ^c	7.06 ± 0.16	7.35 ± 0.14	0.38 ± 0.08 ^b	92.66 ± 0.13 ^c
15	17.21 ± 0.14 ^b	5.56 ± 0.11 ^c	43.45 ± 0.13 ^a	111.45 ± 0.18 ^a	6.57 ± 0.12 ^b	7.09 ± 0.21	7.24 ± 0.15	0.45 ± 0.02 ^a	93.51 ± 0.12 ^c
30	17.41 ± 0.13 ^b	6.07 ± 0.12 ^b	44.06 ± 0.14 ^a	106.79 ± 0.19 ^a	6.77 ± 0.12 ^b	7.14 ± 0.22	7.21 ± 0.11	0.46 ± 0.03 ^a	96.01 ± 0.13 ^b
Effect									
LV ²	0.04	0.05	0.05	0.03	0.05	0.64	0.79	0.05	0.05
TQ ²	0.05	0.05	0.05	0.04	0.05	0.20	0.06	0.01	0.05
TM ²	0.05	0.05	0.05	0.04	0.05	0.19	0.65	0.05	0.05
LV*TQ ³	0.89	0.48	0.93	0.54	0.49	0.08	0.20	0.69	0.41
LV*TM ³	0.73	0.33	0.19	0.66	0.86	0.78	0.81	0.15	0.30
TQ*TM ³	0.47	0.30	0.14	0.25	0.26	0.54	0.64	0.12	0.30
LV*TQ*TM ³	0.65	0.60	0.59	0.65	0.79	0.49	0.52	0.23	0.63
CV, %	6.29	13.81	4.87	12.46	7.04	1.16	1.77	13.06	3.82

¹YH: Yolk height (mm); AH: Albumen height (mm); YD: Yolk diameter (mm); AD: Albumen diameter (mm); YC: Yolk color; YPh: Yolk pH; APh: Albumen pH; ST: Eggshell thickness (µm); HU: Haugh unit; ²means followed by different letters in the column differ by the Tukey test ($p \leq .05$); ³a $p \leq .05$ indicates a direct interaction between the evaluated factors; CV: coefficient of variation.

Table 3. Bacteriological quality of eggs submitted to the BBAO with different levels of AO, and applied using different methods and times.

Factors	Variables ¹		
	Mesophiles	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>
AO level (LV)			
Positive control	0.00 ± 0.00 ^c	0.00 ± 0.00 ^c	0.00 ± 0.00
Negative control	6.69 ± 0.12 ^a	5.27 ± 0.12 ^b	0.27 ± 0.01
1	5.78 ± 0.13 ^a	9.12 ± 0.09 ^a	0.08 ± 0.01
5	1.64 ± 0.12 ^b	2.17 ± 0.11 ^c	0.01 ± 0.00
10	0.55 ± 0.12 ^c	0.00 ± 0.00 ^c	0.00 ± 0.00
15	0.29 ± 0.05 ^c	1.77 ± 0.11 ^c	0.00 ± 0.00
Technique (TQ)			
Positive control	0.00 ± 0.00 ^c	0.00 ± 0.00 ^c	0.00 ± 0.00
Negative control	6.69 ± 0.12 ^a	5.27 ± 0.12 ^a	0.27 ± 0.01
Spray	3.25 ± 0.13 ^b	1.09 ± 0.02 ^b	0.04 ± 0.01
Dip	0.88 ± 0.05 ^c	5.45 ± 0.13 ^a	0.00 ± 0.00
Time (TM)			
Positive control	0.00 ± 0.00 ^c	0.00 ± 0.00 ^c	0.00 ± 0.00
Negative control	6.69 ± 0.12 ^a	5.27 ± 0.12 ^a	0.27 ± 0.01
15	3.35 ± 0.10 ^b	4.50 ± 0.11 ^a	0.00 ± 0.00
30	0.78 ± 0.09 ^c	2.03 ± 0.12 ^b	0.04 ± 0.01
Effect			
LV ²	0.05	0.04	0.58
TQ ²	0.05	0.05	0.56
TM ²	0.05	0.05	0.56
LV*TQ ³	0.41	0.05	0.45
LV*TM ³	0.42	0.70	0.45
TQ*TM ³	0.22	0.93	0.30
LV*TQ*TM ³	0.42	0.45	0.44
CV ⁴ , %	2.84	2.26	3.92

¹All presented values should be multiplied by 10³; ²means followed by different letters in the column differ by the Tukey test ($p \leq .05$); ³a $p \leq .05$ indicates a direct interaction between the evaluated factors; CV: coefficient of variation.

significant ($p \leq .05$) conservation of fat content in eggs stored for 7 days at room temperature compared to those stored without BBAO (negative control). Despite the BBAO coating, there was a significant ($p \leq .05$) reduction in protein content in these eggs compared to control treatments. This reduction in protein content was consistent, regardless of the BBAO technique ($p \leq .05$) or application duration ($p \leq .05$). Additionally, moisture content increased ($p \leq .05$) in eggs treated with BBAO, particularly when the dipping technique was used, with no significant differences in application time.

The protective barrier formed by the BBAO influenced the eggs' chemical composition, mainly by preserving yolk lipids due to the hydrophobic properties of lipid-based coatings that prevent moisture loss and oxygen penetration (Cindric et al., 2007). This protection minimizes yolk's lipid oxidation, although prolonged storage may still promote albumen fluidization and protein denaturation (Pissinati et al., 2014; Rêgo et al., 2012). As the levels of AO in the BBAO increased, yolk lipid oxidation significantly decreased ($p \leq .05$), with higher AO concentrations yielding oxidation levels similar to those of fresh eggs, confirming the efficiency of BBAO in maintaining yolk quality during storage. Coatings based on lipids create a hydrophobic shield that effectively prevents moisture loss and the infiltration of external oxygen into the egg (Pissinati et al., 2014; Salgado et al., 2018), playing a crucial role in safeguarding the lipids predominantly found in egg yolks and contributing to the reduction of lipid oxidation activity during storage (Salgado et al., 2018). Consequently, eggs treated with BBAO, especially those with higher AO levels, exhibited lower yolk lipid oxidation comparable to freshly collected eggs, confirming that this protective

Table 4. Chemical composition and lipid oxidation of the yolks of eggs submitted to the BBAO with different levels of AO, and applied using different methods and times.

Factors	Variables				
	Moisture ¹	Minerals ¹	Fats ¹	Proteins ¹	TBARS values ²
AO level (LV)					
Positive control	75.12 ± 0.15	0.82 ± 0.22	10.94 ± 0.15 ^a	13.12 ± 0.22 ^a	0.170 ± 0.05 ^c
Negative control	77.12 ± 0.19	0.89 ± 0.13	9.64 ± 0.26 ^b	12.35 ± 0.29 ^b	0.242 ± 0.09 ^a
1	77.43 ± 0.25	0.82 ± 0.21	10.42 ± 0.13 ^a	11.33 ± 0.23 ^c	0.221 ± 0.19 ^b
5	77.51 ± 0.29	0.78 ± 0.13	10.70 ± 0.26 ^a	11.01 ± 0.21 ^c	0.216 ± 0.09 ^b
10	77.33 ± 0.27	0.77 ± 0.18	10.71 ± 0.21 ^a	11.19 ± 0.20 ^c	0.206 ± 0.05 ^b
15	77.23 ± 0.31	0.82 ± 0.17	10.47 ± 0.22 ^a	11.48 ± 0.20 ^c	0.196 ± 0.08 ^{bc}
Technique (TQ)					
Positive control	75.12 ± 0.15 ^c	0.82 ± 0.22	10.94 ± 0.15	13.12 ± 0.22 ^a	0.170 ± 0.05 ^c
Negative control	75.09 ± 0.19 ^c	0.89 ± 0.13	11.67 ± 0.26	12.35 ± 0.29 ^b	0.242 ± 0.09 ^a
Spray	77.12 ± 0.23 ^b	0.82 ± 0.17	10.45 ± 0.23	11.61 ± 0.26 ^c	0.215 ± 0.08 ^b
Dip	78.37 ± 0.25 ^a	0.78 ± 0.18	9.96 ± 0.23	10.89 ± 0.25 ^c	0.204 ± 0.06 ^b
Time (TM)					
Positive control	75.12 ± 0.15 ^b	0.82 ± 0.22	10.94 ± 0.15	13.12 ± 0.22 ^a	0.170 ± 0.05 ^c
Negative control	75.09 ± 0.19 ^b	0.89 ± 0.13	11.67 ± 0.26	12.35 ± 0.29 ^b	0.242 ± 0.09 ^a
15	77.58 ± 0.26 ^a	0.81 ± 0.18	10.31 ± 0.21	11.30 ± 0.22 ^c	0.210 ± 0.09 ^b
30	77.92 ± 0.29 ^a	0.79 ± 0.25	10.09 ± 0.25	11.20 ± 0.23 ^c	0.208 ± 0.08 ^b
Effect					
LV ³	0.14	0.66	0.05	0.05	0.01
TQ ³	0.01	0.38	0.14	0.01	0.03
TM ³	0.05	0.64	0.42	0.05	0.01
LV*TQ ⁴	0.16	0.65	0.73	0.29	0.79
LV*TM ⁴	0.53	0.08	0.34	0.23	0.99
TQ*TM ⁴	0.42	0.60	0.88	0.83	0.65
LV*TQ*TM ⁴	0.94	0.93	0.92	0.93	0.95
CV ⁵ , %	1.47	7.02	6.01	7.27	7.12

¹All presented values are expressed in %; ²all values presented are expressed in mgTMP/kg; ³means followed by different letters in the column differ by the Tukey test ($p \leq .05$); ⁴a $p \leq .05$ indicates a direct interaction between the evaluated factors; CV: coefficient of variation.

mechanism effectively conserves the quality and freshness of egg lipids and making BBAO an efficient tool in mitigating lipid oxidation during storage.

3.4 Sensory analysis

The sensory analysis of the eggs, as presented in Table 5, revealed a clear linear trend ($p \leq .05$) in reduced sensory acceptance as the AO level within the BBAO increased. Eggs dipped in the BBAO exhibited significantly higher sensory acceptance ($p \leq .05$) for aroma, color, appearance, and flavor according to the judges, in contrast to eggs treated with spraying or from the negative control. Furthermore, prolonging the exposure time of eggs to BBAO resulted in a notable decline ($p \leq .05$) in sensory acceptance, as evaluated by the judges.

Eggs treated with BBAO containing higher levels of AO exhibited compromised sensory attributes compared to those from the experimental controls. This decline could be attributed to two factors: the transfer of organoleptic properties from AO to the eggs and/or the influence of the BBAO film on the egg's internal content interactions (Biladeau & Keener, 2009; Eddin et al., 2019). The application technique also played a crucial role in sensory outcomes. Eggs treated through immersion received significantly higher sensory acceptance scores for aroma, color,

appearance, and flavor, while eggs subjected to the spray method or from the negative control had comparatively lower scores. This suggests the superiority of the immersion technique in maintaining or enhancing sensory qualities (Pissinatti et al., 2014). Prolonged exposure to BBAO led to a significant reduction in sensory acceptance, indicating the potential adverse effects of extended contact (Pires et al., 2015; Eddin et al., 2019).

4 CONCLUSION

The BBAO effectively preserved the physical, bacteriological, and chemical qualities of eggs stored at room temperature for 7 days, with higher concentrations of AO improving key parameters of physical quality. The immersion technique, especially using 15% AO for 30 s, provided the best preservation results, forming a protective barrier. BBAO significantly reduced microbial contamination, eliminating or minimizing bacterial concentrations. Additionally, the coating lowered lipid oxidation and maintained yolk lipid content, confirming AO's effectiveness as a natural preservative that protects against oxygen exposure and oxidative degradation in eggs at room temperature. However, a decrease in sensory acceptance was observed when higher concentrations of AO were used.

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Table 5. Sensory quality of eggs submitted to the BBAO with different levels of AO, and applied using different methods and times.

Factors	Variables			
	Aroma	Color	Appearance	Taste
AO level (LV)				
Positive control	6.50 ± 0.15 ^a	6.60 ± 0.16 ^a	6.80 ± 0.26 ^a	7.50 ± 0.23 ^a
Negative control	5.10 ± 0.06 ^c	6.20 ± 0.09 ^b	5.80 ± 0.18 ^b	5.70 ± 0.20 ^c
1	5.77 ± 0.15 ^b	5.72 ± 0.18 ^b	5.25 ± 0.22 ^b	6.52 ± 0.15 ^b
5	5.17 ± 0.08 ^{bc}	5.70 ± 0.29 ^b	5.45 ± 0.08 ^b	6.20 ± 0.29 ^b
10	5.35 ± 0.13 ^b	5.67 ± 0.25 ^b	5.72 ± 0.23 ^b	6.07 ± 0.20 ^b
15	5.07 ± 0.05 ^c	5.00 ± 0.23 ^c	4.50 ± 0.25 ^c	5.27 ± 0.18 ^c
Technique (TQ)				
Positive control	6.50 ± 0.15 ^a	6.60 ± 0.16 ^a	6.80 ± 0.26 ^a	7.50 ± 0.23 ^a
Negative control	5.10 ± 0.06 ^c	6.20 ± 0.09 ^b	5.80 ± 0.18 ^b	5.70 ± 0.20 ^c
Spray	5.16 ± 0.08 ^c	5.18 ± 0.09 ^c	4.67 ± 0.26 ^c	6.12 ± 0.21 ^b
Dip	5.52 ± 0.19 ^b	5.86 ± 0.24 ^b	5.78 ± 0.23 ^b	5.91 ± 0.23 ^b
Time (TM)				
Positive control	6.50 ± 0.15 ^a	6.60 ± 0.16 ^a	6.80 ± 0.26 ^a	7.50 ± 0.23 ^a
Negative control	5.10 ± 0.06 ^c	6.20 ± 0.09 ^b	5.80 ± 0.18 ^b	5.70 ± 0.20 ^c
15	5.52 ± 0.19 ^b	5.64 ± 0.17 ^b	5.49 ± 0.08 ^c	6.16 ± 0.25 ^b
30	5.16 ± 0.08 ^c	5.41 ± 0.13 ^b	4.97 ± 0.19 ^c	5.87 ± 0.26 ^c
Effect				
LV ¹	0.04	0.05	0.02	0.02
TQ ¹	0.05	0.02	0.01	0.05
TM ¹	0.05	0.05	0.04	0.05
LV*TQ ²	0.98	0.84	0.57	0.47
LV*TM ²	0.50	0.57	0.39	0.73
TQ*TM ²	0.72	0.76	0.25	0.12
LV*TQ*TM ²	0.83	0.90	0.91	0.88
CV ³ , %	3.62	3.80	4.16	3.60

¹Means followed by different letters in the column differ by the Tukey test ($p \leq .05$); ²a $p \leq .05$ indicates a direct interaction between the evaluated factors; CV: coefficient of variation.

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