Characterization of chilled pasteurized egg white with chocolate flavor

Caracterização de clara de ovo pasteurizada resfriada com sabor chocolate

DOI: 10.55905/oelv21n12-114

Recebimento dos originais: 11/11/2023
Aceitação para publicação: 11/12/2023

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ABSTRACT
Egg products are widely used in the food industry, mainly for manufacturing products aimed at people who practice physical activity. Among them, egg white, in addition to being very versatile, has nutritional benefits for a protein product offering all the essential amino acids in addition to health benefits. Thus, the egg industries seek other ways of presenting this ready-to-eat product, one of them being the addition of flavor, in order to present a more pleasant sensorial product. Thus, this study aimed to characterize physicochemically and microbiologically the pasteurized egg white with chocolate flavor. For this, analyzes of pH, color, nutritional composition, water activity, microbiology and texture were carried out, as well as sensory analysis to evaluate the acceptance of the product. Finally, it was concluded that the chocolate-flavored egg white is a good flavor
option for the product, presenting characteristics very close to the traditional one, maintaining the benefits and adding others, such as masking the darkening caused by processing and promoting a more pleasant flavor to consumption.

**Keywords:** healthy food, protein drink, shelf life.

**RESUMO**
Ovoprodutos são amplamente utilizados na indústria de alimentos, principalmente para a fabricação de produtos destinados a pessoas que praticam atividade física. Entre eles, a clara de ovo, além de ser muito versátil, tem benefícios nutricionais para um produto proteico que oferece todos os aminoácidos essenciais, além de benefícios à saúde. Assim, as indústrias de ovos buscam outras formas de apresentar esse produto pronto para consumo, uma delas sendo a adição de sabor, para apresentar um produto sensorial mais agradável. Assim, este estudo teve como objetivo caracterizar físico-química e microbiologicamente a clara de ovo pasteurizada com sabor a chocolate. Para isso, foram realizadas análises de pH, cor, composição nutricional, atividade hídrica, microbiologia e textura, além de análise sensorial para avaliar a aceitação do produto. Por último, concluiu-se que a clara de ovo com sabor a chocolate é uma boa opção de sabor para o produto, apresentando características muito próximas das tradicionais, mantendo os benefícios e acrescentando outros, como mascarar o escurecimento causado pela transformação e promover um sabor mais agradável ao consumo.

**Palavras-chave:** alimentos saudáveis, bebida proteica, vida útil.

**1 INTRODUCTION**

Egg is a very complete and versatile food, being one of the few foods used worldwide (Silva et al., 2016). Egg products are eggs removed from their shells and processed, which can be the whole egg or just the egg white and yolk separately, arranged in liquid, frozen or dehydrated form (Atilgan et al., 2008). More and more consumers are looking for products in supermarkets that are healthy, practical and have a pleasant flavor. The high use of eggs as raw material is due to their low cost and health benefits, thus producing differentiated products with high added value (Shrestha et al., 2023; Sun et al., 2023, Wu et al., 2024, Zang et al., 2023).

Although egg products are widely used industrially, only cooled pasteurized whole egg and dehydrated egg have identity and quality standards defined by the Ministry of Agriculture (Brasil, 1991).
Ovalbumin and other egg white proteins are considered a good source of amino acids, and egg white has the 8 essential amino acids that are very important in the diet of athletes to build and also maintain muscle mass (Lotfian et al., 2019a). Furthermore, the bioavailability of egg protein is very high when compared to other animal protein sources (Silva et al., 2016).

Proteins have numerous nutritional functions in the body, contributing essential structural components, as well as providing the basic materials for hormones and supporting immunity. They are used in different ways throughout the body, depending on their source, it is important to ingest proteins that can properly contribute to muscle health (Matsuoka et al., 2017).

To meet industrial demand, egg products undergo pasteurization for liquid use or dehydration for powder use. In this way, they show several benefits, such as microbiological safety, recipe standardization, storage space, longer shelf life and more practicality (Badr, 2006).

One of the functional foods that athletes look for to build muscle and gain athletic strength are ready-to-drink protein drinks, which are often developed based on milk, egg or soy proteins due to their high nutritional and functional benefits compared to other sources (Lotfian et al., 2019b).

In addition to consumption by athletes, Jiayu et al. (2020) demonstrated that regular consumption of egg whites can help prevent diseases such as cardiovascular diseases, while Jiang et al. (2021) studied the benefits of egg white consumption for the liver fat, where they observed that the consumption of powdered egg white helps in the reduction of hepatic triacylglycerol. Furthermore it is still an excellent candidate for the maintenance of physiological homeostasis or, in other words, it helps to maintain the functions that the organism needs for the balance of the body (Song et al., 2014).

According to Oma et al. (2021), the study of new technologies for the application of thermal/non-thermal treatments in dehydrated egg whites has increased and has shown satisfactory results, but, on a laboratory scale only. However, despite the good results and application to the industry, the economic feasibility for application on an industrial scale,
as well as sensory evaluation and addition of flavorings in products were not raised, which would be interesting for future works.

It is also important to emphasize that it is not only the industrial process that causes losses in the functionality of the egg white, for, since the egg raw, its functions can undergo changes according to the packaging temperature and also to the storage time (Liu et al. 2018). During the storage period, the egg whites can undergo physicochemical changes that can be intensified depending, mainly, on temperature and relative humidity. These changes involve pH, viscosity and modifications in their proteins and, consequently, impact their functionality (Santana and Muller, 2016).

Knowing the variables that affect the performance of proteins that give egg whites so many functions and benefits to their consumption, as well as knowing and developing ways to avoid such changes can bring great benefits to the egg industry, final consumers and the food industries that use egg products in their formulations, by obtaining egg products even more similar to the product raw, but with greater safety and other practicalities already mentioned above. As far as is known, this work is pioneering and presents an innovative character for evaluating the use of chocolate flavoring in egg whites.

Therefore and due to the growing search of consumers for industrialized, healthy and practical foods, this work aimed to characterize the nutritional composition, physicochemical and microbiology of cooled pasteurized egg white chocolate flavor, as well as to evaluate its sensory acceptance.

2 MATERIAL AND METHODS

2.1 OBTAINING PASTEURIZED EGG WHITES

The traditional cooled pasteurized egg white and chocolate flavor were processed in industrial equipment and supplied by an egg industry in the city of Iacri/SP. They were pasteurized at 56.7°C for 3.5 minutes, being immediately cooled to a maximum of 5°C.

Two samples were obtained, being traditional pasteurized egg whites (CL) and pasteurized egg white with chocolate flavored (CLC) and the only difference between them was the addition of cocoa, chocolate flavor and synthetic sweetener, which make up
less than 2% of the product’s formulation. The evaluated batch consisted of 24 samples of 1 kg of each flavor, being randomized for storage and analysis.

2.2 MICROBIOLOGY

The samples of pasteurized egg white with chocolate flavored were analyzed (on the day after manufacture and also at the end of their shelf life) microbiologically for: Total mesophilic count (CFU/g) according to ISO 4833-1:2015 method, Coliforms total and thermotolerant (MPN/g) according to ISO 4832:2010, S. aureus (/g) according to ISO 6888-1: 1999, Salmonella spp (/25g) according to the American Public Health Association method (1978) and Enterobacteriaceae (CFU/g) according to ISO 21528-2:2004.

2.3 CHEMICAL COMPOSITION AND pH

The chemical composition analysis was performed in an external laboratory (Hidrolabor) in the city of Sorocaba/SP for protein content (ISO 1871:2009), total fat (AOAC 925.32: 2019) and ash (Method 4.4 of the Manual of Official Methods for Analyzing Foods of Animal Origin) (Brasil, 2018). Moisture analysis was performed according to AOAC (2004) in an oven at 105°C until constant weight. The amount of carbohydrates was calculated by difference, according to equation 1 (Lovato et al., 2018).

\[
\text{Total Carbohydrates} = 100 - (\text{water} + \text{protein} + \text{total fat} + \text{ash}) \, (\%)
\] (equation 1)

The pH was measured using a Gehaka model PG 1800 pH meter, introducing the electrode directly into the sample. The samples were stored in a carton package at a temperature of 5°C ± 1°C and 35°C ± 1°C and the pH was measured weekly until the identification of a change in its standard pH (until the end of its shelf life).

A part of the samples was stored in an incubator at 35°C ± 1°C and the pH was evaluated weekly for chocolate-flavored egg whites until a change was identified.
2.4 WATER ACTIVITY

The water activity analysis was performed using the equipment AQUALAB® 4TE Water Activity Meter, in which, firstly, the measurement was performed in distilled water for calibration and, then, the measurement was performed on the samples in triplicate.

2.5 COLOR ANALYSIS

The color of each sample was analyzed using a portable Minolta ® CR400 colorimeter, with an integration sphere and a viewing angle of 3° (illumination d/3 and illuminant D65). The sample was placed directly from the 1kg carton package, evenly, in a 90 x 15mm petri dish, with each analysis performed in triplicate. The color was determined with the CIELAB system (L* representing the luminosity - from dark (0) to light (100), the a* coordinate representing the green (-) and red (+) and the b*- coordinate representing the blue (-) and yellow (+). The total color difference (ΔE) was employed using the equation (2) (Katekhong et al., 2017).

\[ \Delta E = \sqrt{(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2} \] (equation 2)

Where L*0, a*0 e b*0 are the sample color values at time zero and L*1, a*1 e b*1 are the values of the sample taken at time t. Since high values of ΔE indicate large color changes in relation to the reference.

2.6 SENSORY EVALUATIONS

The attributes acceptance test (aroma, color, flavor and global acceptance) was carried out with 60 volunteer, physical activity practitioners, untrained, male and female tasters with no predetermined age. A nine-point hedonic scale was used (1 = I disliked it very much and 9 = I liked it very much) and a survey with 3 questions about the consumption of egg whites. The panelists received approximately 20 mL of each sample, at a temperature varying around 5°C, in disposable plastic cups with a capacity of 50 mL, coded with three-digit random numbers.
With the results obtained, the acceptance index was calculated according to the equation (3) (Ribeiro et al., 2020).

\[ IA = \left( \frac{A}{B} \right) \times 100 \]  

(equation 3)

Where, A represents the average grade received by the sample according to the hedonic scale and B represents the highest grade received by the sample according to the hedonic scale.

2.7 STATISTICAL ANALYSIS

Each sample condition was tested in triplicate at each time point. To compare changes in dynamic parameters, ANOVA was performed with Tukey's multiple comparison test (p<0.05).

3 RESULTS AND DISCUSSION

3.1 MICROBIOLOGICAL ANALYSIS

According to the results obtained (Table 1), it shows that the product meets the microbiological standards both at the beginning and at the end of its shelf life, taking into account the standards recommended by IN 60 of December 23, 2019 (Anvisa, 2019) as well as European regulation EC 1141/2007 (Commission Regulation 2007), which require max. 10 CFU/g of *Enterobacteriaceae* and absence of *Salmonella*, the latter being also required by the FSIS (Food Safety and Inspection Service) (2005) of the United States Department of Agriculture (USDA). The other analyzes carried out and presented are analyzes adopted by the industry, since they are indicators of good manufacturing practices.
Table 1 – Microbiological results for the samples of egg white chocolate-flavored at the beginning and end of their shelf life.

<table>
<thead>
<tr>
<th>Parameter/sample</th>
<th>CLCi</th>
<th>CLCf</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total mesophilic count (CFU/g)</td>
<td>$1.0 \times 10^1$ (1.00 log)</td>
<td>$1.09 \times 10^3$ (3.04 log)</td>
</tr>
<tr>
<td>Total coliform (NMP/g)</td>
<td>&lt;0.3</td>
<td>&lt;0.3</td>
</tr>
<tr>
<td>Fecal coliform (/g)</td>
<td>Absent</td>
<td>Absent</td>
</tr>
<tr>
<td>Staphylococcus aureus (/g)</td>
<td>Absent</td>
<td>Absent</td>
</tr>
<tr>
<td>Salmonella spp (/25g)</td>
<td>Absent</td>
<td>Absent</td>
</tr>
<tr>
<td>Enterobacteriaceae (CFU/g)</td>
<td>Absent</td>
<td>Absent</td>
</tr>
</tbody>
</table>

CLCi: egg white chocolate flavored right after production; CLCf: egg white chocolate flavored at the end of its shelf life.

Source: The Authors (2023).

The results of total count of mesophiles, total coliforms and *S. aureus* are in agreement with the results obtained by Cwiková and Nedomová (2014) who found a value of 3.1 log for mesophiles, while total coliforms were detected in two of their samples and *S. aureus* in only one sample.

Badr (2006) when applying irradiation in chilled egg white liquid found, at time zero, a count of 2.99 log in the analysis of total mesophilic count, not detecting *Enterobacteriaceae* neither *S. aureus* and a negative result for *Salmonella*, which is also in agreement with the results obtained.

These results demonstrate that egg white chocolate flavored is microbiologically safe for consumption and meet national and international food safety standards.

3.2 ANALYSIS OF CHEMICAL COMPOSITION, pH, TEXTURE AND AW

Although there is no current Brazilian legislation, the industry adopts min. 10g/100g for protein, max. 0.5g/100g for total fat, max. 0.7g/100g for fixed mineral residue and max. 1.1g/100g for carbohydrates that are very close to that found in the USDA (2019) nutritional table for egg whites, where for 100g of egg whites 10.9g of protein, 0.17g of fat, 0.73g of carbohydrates and 0.63g of ash. Thus, the results obtained for the egg whites are presented in table 2.
Table 2 – Chemical composition of traditional egg white and chocolate flavor.

<table>
<thead>
<tr>
<th>Parameter/sample</th>
<th>CL</th>
<th>CLC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein (g/100g)</td>
<td>9.92±0.05</td>
<td>9.64±0.09</td>
</tr>
<tr>
<td>Moisture (g/100g)</td>
<td>88.96±0.04</td>
<td>88.16±0.01</td>
</tr>
<tr>
<td>Total fat (g/100g)</td>
<td>0.34±0.04</td>
<td>0.35±0.09</td>
</tr>
<tr>
<td>Ash (g/100g)</td>
<td>0.67±0.07</td>
<td>0.74±0.05</td>
</tr>
<tr>
<td>Carbohydrates (g/100g)</td>
<td>0.11±0.05</td>
<td>1.12±0.19</td>
</tr>
</tbody>
</table>

* Equal letters on the same line represent statistically equal results by the Tukey test (p<0.05).
Source: The Authors (2023).

Lechevalier et al. (2005) evaluated the dry matter of chilled pasteurized egg white obtaining a value of 112 g/kg (or 11.2g/100g), which can be expressed on a wet basis as 88.8 g/100g. In the study by Santana and Muller (2016), values of 87% moisture and 11% protein were obtained for pasteurized egg whites. The aforementioned data in the two studies corroborate the data obtained in the present research (Table 2).

From this correlation, it is possible to compare the results obtained for both CL and CLC, since they did not present a significant difference (p≤0.05), and observe the similarity between them.

The results of chemical composition analyzes show a lot of similarity between CL and CLC, differing only in the amount of carbohydrates and moisture (p<0.05), probably due to the addition of the ingredients.

There are few works found in the literature on the addition of flavorings in egg whites. In the study by Lotfian et al. (2019b), the protein drink added to dehydrated egg white showed a significant increase in protein and a reduction in fats and carbohydrates, indicating the high concentration of protein in the egg white. Canela-Rawls (2014) shows that the egg white is composed of 88% water, 11% protein, 0.2% fat and 0.8% minerals.

Thus, the results of the nutritional composition obtained in this work are in accordance with both the literature and other works for traditional egg whites and characterize the egg whites chocolate flavored nutritionally very similar to the traditional one, differing only in the carbohydrate content.

The pH values were monitored in the CL and CLC samples before and after the pasteurization process as well as before and after the addition of ingredients in the case of egg white with chocolate flavor (Table 3). Note that the pH values obtained for CL are in agreement with the results obtained by Lechevalier et al. (2005), who found a value of
9.15 ± 0.05. The work by Badr (2006) showed a pH of 8.47 for liquid egg white before and after the thermal process and keeping it under refrigeration, being this value closer to the value found for CLC, this factor can be explained by the variability of the raw material, since it undergoes pH variations during its storage (Santana and Muller, 2016).

Table 3 – pH results.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>No ingredients added</th>
<th>Ingredientes added</th>
<th>After pasteurization</th>
</tr>
</thead>
<tbody>
<tr>
<td>CL</td>
<td>9.76±0.06</td>
<td>-</td>
<td>9.91±0.05</td>
</tr>
<tr>
<td>CLC</td>
<td>9.41±0.01</td>
<td>8.88±0.01</td>
<td>9.44±0.01</td>
</tr>
</tbody>
</table>

* Equal letters on the same line represent statistically equal results by the Tukey test (p<0.05).

Source: The Authors (2023).

No studies were found that point out physicochemical characteristics of protein drinks based only on egg whites. Protein drinks found in studies use a mix of proteins, such as milk and egg whites, like what was found in the study by Lotfian et al. (2019a). The authors observed in the formulation that used the highest percentage of dehydrated egg white (16%), pH values of the drink between 8.75 - 8.20 over 10 days of storage under refrigeration.

The results mentioned above were considered, at first, as comparative, since the composition has a higher percentage of milk (80%), which has a pH close to neutrality, followed by egg white (16%) and cocoa (1%), which will impact the pH of the drink, sugar (2.8%) and carrageenan gum (0.2%). Table 4 shows the results of the textural analyzes carried out on the egg whites at room temperature.

Table 4 – Texture parameters of egg whites at 25°C.

<table>
<thead>
<tr>
<th>Texture/Sample</th>
<th>CL</th>
<th>CLC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Firmness (g)</td>
<td>26.70±0.63</td>
<td>27.63±0.50</td>
</tr>
<tr>
<td>Consistency (g.s)</td>
<td>154.01±1.14</td>
<td>156.73±0.02</td>
</tr>
<tr>
<td>Cohesiveness (g)</td>
<td>-5.99±0.19</td>
<td>-5.85±0.27</td>
</tr>
<tr>
<td>Viscosity Index (g.s)</td>
<td>-0.84±0.05</td>
<td>-0.85±0.08</td>
</tr>
</tbody>
</table>

* Equal letters on the same line represent statistically equal results by the Tukey test (p<0.05).

Source: The Authors (2023).

According to the results presented, it is possible to verify that no significant difference was observed for any of the parameters, which shows that the addition of the
ingredients did not impact the textural parameters of the egg white with chocolate flavored.

In order to characterize the egg whites, the water activity was also analyzed, and both had an average value of 1.04 with a standard deviation of 0.002 and 0.004 for traditional egg white and chocolate flavored, respectively. This result does not show a significant difference (p<0.05) between the samples, being another parameter that was not impacted by the addition of ingredients.

3.3 COLOR ANALYSIS

The color values (L, a and b) of egg whites chocolate-flavored during storage at 5°C ± 1°C are shown in table 5.

<table>
<thead>
<tr>
<th>Days</th>
<th>L*</th>
<th>a*</th>
<th>b*</th>
<th>∆E</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>19.60±0.15</td>
<td>10.66±0.44</td>
<td>9.07±0.32</td>
<td>-</td>
</tr>
<tr>
<td>14</td>
<td>21.01±0.92</td>
<td>10.69±0.42</td>
<td>15.13±0.45</td>
<td>6.33±0.50</td>
</tr>
<tr>
<td>44</td>
<td>20.36±0.25</td>
<td>11.73±0.59</td>
<td>16.80±0.10</td>
<td>7.63±0.05</td>
</tr>
<tr>
<td>75</td>
<td>25.50±0.45</td>
<td>11.88±0.15</td>
<td>17.65±0.33</td>
<td>6.41±0.34</td>
</tr>
</tbody>
</table>

Source: The Authors (2023).

It is noted that at the end of storage, it seems, the samples were lighter and with a greater tendency to red and yellow, with a color variation (ΔE) of 6.41. Obón et al. (2009) cites that color changes can be measured as the magnitude of the distance vector between the initial color values and the actual color coordinates in the CIELAB three-dimensional color space (ΔE). In addition, differences in perceptible color can be defined as small (ΔE < 1.5), distinct (1.5 < ΔE < 3) and highly distinct (ΔE > 3) the author also mentions that the color variation is only evident for ΔE > 3. For egg white with chocolate flavored, no color values were found in other studies, thus making it impossible to make a comparison regarding this parameter.

In the work of Santana and Muller (2016) it was observed that pasteurized egg white undergoes darkening throughout its shelf life, starting with its industrial processing.
Therefore, it can be said that the addition of ingredients such as those used in the egg white with chocolate flavor helps to mask this darkening, being a benefit to the industry.

3.4 SENSORY ANALYSIS

As for the consumption of egg white, 71.66% of the panelists consume egg white more than once a week and only 18.33% do not consume it. Among those who consume egg whites, 53.85% consume them as an omelet or in a shake/vitamin and 90.2% purchase the egg whites in their raw form, that is, the whole egg in shell for later separation.

The high percentage of tasters who purchase fresh egg whites (whole egg in shell) may indicate a lack of knowledge or, even, difficulty in finding something to buy, indicating a good market to be explored.

The results obtained in the sensory evaluation indicate that the egg white with chocolate flavored (Figure 1) was better evaluated in all parameters when compared to the traditional one (Figure 2), which is also confirmed by the acceptance rate shown in Figure 3.

Figure 1 – spider chart of sensory analysis of egg white chocolate flavored.

![Sensory - CLC](source: The Authors (2023).)
Sensory analysis was performed on the work of Marinha (2015) of egg white in the flavors of strawberry, vanilla, banana, chocolate and traditional. In this study, a better evaluation was also verified for all flavors compared to the traditional one. Chocolate was
not the best flavor evaluated, however, it underwent texture changes due to the composition tested. Still, the author also reported that there was a lot of variation in the results justified by the personal tastes of the tasters.

Finally, it is noteworthy that the acceptance rate for egg white with chocolate flavored presented results above 87% for all parameters, indicating a good flavor option, which would facilitate and encourage the consumption of this type of product.

4 CONCLUSIONS

With the results obtained, it was possible to characterize microbiologically the cooled pasteurized egg white with chocolate flavored, as well as to evaluate its sensory acceptance, where it obtained a high acceptance rate (IA>87%), in addition to presenting itself as a safe and more pleasant product, sensorially, to consumption. The addition of flavoring in general did not influence the proximate composition, aw or textural parameters of the samples evaluated.

According to the color monitoring, it was concluded that the addition of inputs gave color to the product, masking the darkening that the traditional egg white undergoes in processing and throughout its shelf life.

Finally, it is concluded that egg white with chocolate flavored is a safe and pleasant option of taste for a great source of proteins, and, when talking about flavored egg whites, there are still fields to be explored, such as the impact of ingredients on its shelf life and its applicability in recipes with relatively simple processing and with a growing public looking for products with the characteristics obtained, in addition to the high added value of the final product, showing a high profitability for the industry.

CONFLICT OF INTEREST

Authors declare that they have no conflict of interest.
REFERENCES


ISO 6888-1:1999. Microbiology of food and animal feeding stuffs - Horizontal method for the enumeration of *Staphylococcus aureus*.


ISO 4833-1:2015. Microbiology of food and animal feeding stuffs - Horizontal method for the enumeration of micro-organisms, Colony-count technique at 30 °C.


