SCIENTIA PLENA
www.scientiaplena.org.br
VOL. 21, NUM. 01 2025
doi: 10.14808/sci.plena.2025.011501

The role of culinary herbs on the thermal degradation of carotenoids in omelets prepared by different methods

O papel de ervas culinárias na degradação térmica de carotenóides em omeletes preparadas por diferentes métodos

V. S. de Oliveira[1*]; D. W. H. Chávez[1]; I. M. Augusta[1]; O. D. Gamallo[1]; R. N. Castro[2]; T. Saldanha[1]

[1]*Department of Food Technology/Institute of Technology, Federal Rural University of Rio de Janeiro, 23890-000, Seropédica-Rio de Janeiro, Brazil*
[2]*Department of Organic Chemistry/Institute of Chemistry, Federal Rural University of Rio de Janeiro, 23890-00, Seropédica-Rio de Janeiro, Brazil*

**vanessasdo@bol.com.br*
(Recebido em 09 de dezembro de 2024; aceito em 24 de janeiro de 2025)

This study investigated the effects of adding basil (*Ocimum basilicum* L.), oregano (*Origanum vulgare*), and parsley (*Petroselinum crispum*) on the total carotenoid content of air-fried, microwaved, and pan-fried omelets. The herb extracts were first characterized for their antioxidant capacity and total phenolic and carotenoid contents. Omelets were prepared without herbs (control) and with the herbs incorporated into the egg white and yolk mixture at three levels (0.25%, 0.5%, and 0.75%). In the control samples, cooking significantly reduced carotenoid content, except for pan frying. Air frying caused the greatest reduction, decreasing carotenoids from 14.87 ± 0.09 μg/g (raw) to 10.06 ± 0.11 μg/g, representing a 32.35% degradation. Microwaving resulted in a 23.64% loss. In contrast, higher levels of carotenoids were found in samples containing the herbs, mainly when oregano was used at 0.75%. Overall, the results indicate that while some carotenoids from the herbs may degrade during cooking, their addition can effectively protect egg carotenoids from thermal degradation by acting as natural antioxidants.
Keywords: eggs, oxidation, carotenoids.

Este estudo investigou o efeito da adição de manjericão (*Ocimum basilicum* L.), orégano (*Origanum vulgare)* e salsa (*Petroselinum crispum*) no teor total de carotenóides de omeletes preparadas em *air fryer*, micro-ondas e frigideira. Os extratos de ervas foram inicialmente caracterizados por sua capacidade antioxidante e teores de fenólicos e carotenóides totais. As omeletes foram preparadas sem ervas (controle) e com as ervas incorporadas à mistura de clara de ovo e gema em três níveis (0,25%, 0,5% e 0,75%). Nas amostras de controle, o cozimento reduziu significativamente o teor de carotenóides, exceto para o preparo na frigideira. O preparo em *air fryer* causou a maior redução, diminuindo os carotenoides de 14,87 ± 0,09 μg/g (cru) para 10,06 ± 0,11 μg/g, representando uma degradação de 32,35%. O micro-ondas resultou em uma perda de 23,64%. Em contraste, níveis mais elevados de carotenóides foram encontrados nas amostras contendo as ervas, principalmente quando o orégano foi usado a 0,75%. No geral, os resultados indicam que, embora alguns carotenóides das ervas possam degradar durante o cozimento, sua adição pode efetivamente proteger os carotenóides do ovo da degradação térmica, agindo como antioxidantes naturais.
Palavras-chave: ovos, oxidação, carotenóides.

1. INTRODUCTION

Chicken eggs are an economically affordable source of nutrients and crucial components of a balanced diet. They have significant value for daily meals and serve as basic ingredients in various culinary dishes [1, 2]. Additionally, eggs provide many desirable technological properties and are extensively used by the food industry. As a result, eggs rank among the most widely consumed staple foods worldwide [1, 3].

In addition to being rich in high-quality proteins, lipids, vitamins, and minerals, eggs are an important source of carotenoids [4, 5]. The carotenoids found in egg yolk are highly bioavailable and exhibit a range of beneficial biological activities. Their consumption has been linked to a reduced risk of ocular, cardiovascular, and degenerative diseases, as well as diabetes and certain

cancers [5-7]. Furthermore, carotenoids impart the yellow to orange coloration of egg yolk, which is highly valued by consumers, and they are recognized for their strong antioxidant properties [8].

Carotenoids are highly reactive due to their system of conjugated double bonds, enabling them to function as antioxidants by trapping free radicals or physically quenching singlet oxygen [9, 10]. However, this reactivity also makes them unstable. Due to their unsaturated structures, carotenoids are easily degraded through oxidation and isomerization when exposed to factors such as heat, light, oxygen, and catalytic agents. Consequently, thermal preparation of eggs can degrade carotenoids, potentially compromising their functionality, bioavailability, and overall quality [10-12].

Eggs are frequently consumed as scrambled eggs, commonly referred to as omelets, which can be prepared using various thermal methods. The traditional method involves frying in a conventional pan, but microwaving and air frying have gained popularity as practical alternatives due to their precise process control and space-saving designs [13, 14]. Air fryers cook food by circulating heated air uniformly inside the fryer chamber, achieving the characteristic color and crispness of fried foods without the use of oil [14, 15]. Microwave heating, a form of dielectric heating, eliminates the need for a heat-transfer medium, enabling faster and more uniform cooking [13, 16].

Culinary herbs are widely used both in traditional cuisine and the food industry to enhance the color, flavor, and aroma. Furthermore, herbs have garnered significant research interest due to their nutritional value and health-promoting properties, which are linked to the presence of antioxidant compounds (e.g. phenolic acids, flavonoids, carotenoids) [17]. Due to these antioxidant properties, incorporating herbs into food preparations can support the human endogenous antioxidant system and protect food components from oxidation [18-20].

In this context, given the nutritional and biological significance of carotenoids in eggs, it is worth investigating the impact of cooking methods on carotenoid stability during omelet preparation, a topic that has not yet been thoroughly explored. Therefore, this study aimed to evaluate the role of basil, oregano, and parsley (0.25, 0.5, and 0.75%) on the thermal degradation of carotenoids in omelets prepared by air frying, microwaving, and pan frying.

2. MATERIAL AND METHODS

2.1 Chemicals

Folin–Ciocalteu reagent, 2,4,6-tris(2-pyridyl)-s-triazine, 1,1-diphenyl-2-picrylhydrazyl, gallic acid, and Trolox were purchased from Sigma-Aldrich (St. Louis, MO, USA). All other chemicals were acquired from Vetec (São Paulo, SP, Brazil).

2.2 Herbs

Fresh basil, oregano, and parsley, approximately 1 kg of each herb, were donated by local producers from Seropédica, Rio de Janeiro, Brazil (latitude 22°76'19.31"S; longitude 43°67'16.99"W). The herbs were authenticated and voucher specimens were deposited in the Herbarium of the Federal Rural University of Rio de Janeiro under numbers 45091 (basil), 37741 (parsley), and 56083 (oregano). The leaves were washed in running water, dried with paper towels, and chopped. Next, they were stored in plastic bags and maintained at 4 °C until analyses or, immediately used.

2.3 Herbs' characterization

2.3.1 Extracts

The extracts were prepared as described by de Oliveira et al. (2022) [21]. Each fresh herb (2 g) was homogenized with 40 mL of an ethanol/water solution (80:20, v/v) at 25 °C under mechanical shaking for 30 min. Then, the mixture was sonicated for 20 min (40 kHz) (Elmasonic P, Elma Schmidbauer GmbH, Singen, Germany) and centrifuged at 18,000 x *g* for 10 min (NI 1813, Nova

Instruments, São Paulo, Brazil). Then, the supernatant was transferred to a volumetric flask of 50 mL and the volume was completed with the extracting solution.

2.3.2 Total phenolic content and in vitro antioxidant capacity

These analyzes were performed following the procedures reported by Mariano et al. (2022) [22]. For total phenolic compounds, 1 mL of extract was mixed with 10 mL of distilled water and 1 mL of the Folin–Ciocalteu reagent diluted in distilled water (1:10, v/v). After 3 min, the mixture was homogenized with 1.5 mL of 10% sodium carbonate solution and allowed to rest for 2 h at 25 °C in the dark [23]. The absorbance was measured at 725 nm with a spectrophotometer (WUv-m51, Weblabor, São Paulo, Brazil) and results were presented as mg gallic acid equivalent (GAE)/g sample.

The *in vitro* antioxidant capacity was determined by the DPPH and FRAP assays. For the DPPH radical scavenging assay, a mixture prepared with the extract (100 μL) and 3.9 mL of a 0.06 mM methanolic DPPH solution was homogenized and left resting for 1 h in the dark at 25 °C [24]. Readings were performed at 517 nm and the DPPH radical scavenging activity was determined as follows: $\%I_{DPPH} = (A_0 - A) / A_0) \times 100$, where A_0 and A are the absorbance of the control (DPPH) and sample, respectively.

The FRAP reagent was prepared by mixing 300 mM sodium acetate buffer (pH 3.6), 10 mM TPTZ (2,4,6-tris(2-pyridyl)-s-triazine) solution in 40 mM hydrochloric acid, and 20 mM ferric chloride in proportions of 10:1:1 (v/v/v). The extract (90 μL) was homogenized with distilled water (270 μL) and the FRAP reagent (2.7 mL). The solution was heated in a water bath at 37 °C for 30 min [25]. Then, it was cooled and the absorbance was measured at 595 nm. The results were expressed as μmol Trolox equivalent (TE)/g.

2.3.3 Total content of carotenoids

A methodology that determines carotenoids and chlorophylls simultaneously and is widely applied the in the literature for plant materials was used[26]. Each fresh herb (2 g) was macerated with calcium carbonate (0.2 g) and an acetone/water solution (7 mL; 80:20, v/v). The mixture was filtered to a volumetric flask (25 mL) and the volume was completed with the solution. Readings were performed at 470 (carotenoids), 647 (chlorophyll *a*), and 663 nm (chlorophyll *b*) and the carotenoid content (μg/g) was determined as follows using equations 1, 2, and 3.

$$\text{Chlorophyll } a \text{ (Ca)} = [12.25\ A663.2 - 2.79\ A646.8] \quad (1)$$

$$\text{Chlorophyll } b \text{ (Cb)} = [21.50\ A646.8 - 5.10\ A663.2] \quad (2)$$

$$\text{Total Carotenoids} = [1000\ A470 - (1.82Ca - 104.96\ Cb)/198] \quad (3)$$

2.4 Omelets

2.4.1 Omelet preparation

Fresh eggs (50 ± 0.5 g) were purchased in a market in Seropédica, Rio de Janeiro, Brazil. The yolk and white of twelve eggs were mixed using a mixer (velocity 1 for 15 s, Ri7000, Walita, São Paulo, Brazil) to obtain a homogenous sample, which was analyzed before cooking (raw sample). Subsequently, the same procedures were performed to prepare the cooked samples.

The omelets were cooked according to each treatment: omelets without herb (control) and omelets containing fresh basil, parsley, or oregano leaves at concentrations of 0.25, 0.5, and 0.75%. The amount of herb used was determined based on previous research [27, 28], which used these herbs as natural antioxidant in eggs. Moreover, informal sensory tests were also performed with the laboratory team to establish herb percentages deemed suitable by consumers based on the size of the prepared omelet (150 ± 0.5 g, made with 3 eggs). As the quantity of herbs used in culinary dishes is largely subjective, tests were conducted using various proportions, with visual evaluation as the primary method of analysis [28].

2.4.2 Thermal process

Due to the lack of standard procedures for preparing omelets, the temperature/time binomial of each thermal treatment was determined by performing informal tests with the laboratory team, mimicking domestic preparations as previously reported [27, 28]. In addition, food safety was also considered and a minimum internal temperature of 70 °C was defined [29]. The temperature was monitored with a thermometer (Incoterm 6132, São Paulo, Brazil) to achieve the minimum value requested.

Air frying was performed using an electric air fryer (RI9225/50, Philips Walita, São Paulo, Brazil) set at 220 °C for 10 min. Pan frying was carried out using a domestic stove (Esmaltec, Ceará, Brazil). The sample was prepared using only a Teflon pan without the addition of oil, being turned over at 1-min intervals, totaling 4 min of cooking. For microwaving, a microwave oven (CCE, São Paulo, Brazil) was used to cook the samples for 3 min (900 W) [27, 28].

Three omelets were prepared for each cooking method. Then, they were combined to obtain a single sample, which was ground and lyophilized.

2.4.3 Total content of carotenoids

The sample (3 g) was mixed with 15 mL of acetone and sonicated for 15 min at 40 kHz. The mixture was filtered using a vacuum glass funnel equipped with a sintered plate. The extraction process was repeated until the sample no longer displayed the distinctive carotenoid coloration. The acetone extract was transferred to a separatory funnel containing 15 mL of petroleum ether and washed with distilled water at least 3 times. The ether extract was filtered through anhydrous sodium sulfate, collected in a volumetric flask (25 mL), and the volume was completed with petroleum ether. The procedures were performed using limited light and readings were performed at 450 nm. The results were calculated according to Equation 4, where A is the absorbance, V is the total extract volume, m is the mass of sample, and $A^{1\%}_{1cm}$ is the absorption coefficient of zeaxanthin in petroleum ether [30].

$$\text{Total carotenoid content } (\mu g/g) = A \times V \times 10^4 / A^{1\%}_{1cm} \times m \quad (4)$$

2.5 Statistical analysis

For herbs comparison, it was applied an one way ANOVA, followed by mean test comparison of Tukey when differences were detected. On the other hand, to evaluate the effect of cooking method, the type of the herb, and the amount of herb, it was applied a three way ANOVA, then, where differences were detected, it was applied a Tukey test for multiple comparison. Finally, the Dunnet test was applied to compare the treatments with the control. A significance level of 5% was considered using the software R.

3. RESULTS AND DISCUSSION

3.1 Herbs' analyses: *in vitro* antioxidant capacity, phenolic compounds, and carotenoids

Oregano exhibited the highest antioxidant capacity against the DPPH radical ($p < 0.05$), with an inhibition of 89.36 ± 0.48 %, followed by basil (37.82 ± 0.09 %) and parsley (13.07 ± 0.64 %) (Table 1). These results align with the ones obtained by other authors [31], who described the highest potential of oregano to inactivate DPPH radicals when compared to basil and parsley. Inactivation values for parsley extracts ranging from 2.16% to 14.26% were determined [32], consistent with the present study, whereas a higher value of 59.21% was also reported [18]. Regarding basil, extracts obtained with ethanol, methanol, and water presented inhibition percentages of 6.86, 9.10, and 6.09%, respectively [33].

Table 1: Total content of phenolics and carotenoids and antioxidant capacities determined by DPPH and FRAP assays of basil, oregano, and parsley extracts.

	Basil	Oregano	Parsley
Total phenolics mg GAE/g	8.00 ± 0.75^{b}	11.21 ± 0.20^{a}	5.12 ± 0.03^{c}
Total carotenoids µg/g	6.18 ± 0.04^{c}	12.95 ± 0.09^{a}	11.76 ± 0.12^{b}
DPPH % Inhibition percentage	37.82 ± 0.09^{b}	89.36 ± 0.48^{a}	13.07 ± 0.64^{c}
FRAP µmol Trolox TE/g	217.36 ± 1.14^{b}	134.55 ± 0.16^{a}	321.11 ± 0.35^{c}

Results presented as mean ± standard deviation, n=3. Different letters in the same row mean statistically different results by the Tukey test ($p < 0.05$).

Regarding FRAP, the antioxidant capacity decreased in the following order: parsley (321.11 ± 0.35 µmol TE/g), basil (217.36 ± 1.14 µmol TE/g), and oregano (134.55 ± 0.16 µmol TE/g) ($p < 0.05$) (Table 1). Hossain et al. (2011) [34] reported FRAP values of 232.93 µmol TE/g for basil, 753.53 µmol TE/g for oregano, and 51.14 µmol TE/g for parsley. The result obtained for basil is in agreement with the present study; however, the authors observed a higher reducing power for oregano and a lower potential for parsley. Lower values, compared to the ones from this study, were also described for oregano (51.56 µmol TE/g) and parsley (5.99 µmol TE/g) [35, 36].

For the total phenolic contents, oregano showed the highest amount (11.21 ± 0.20 mg GAE/g), followed by basil (8.00 ± 0.75 mg GAE/g) and parsley (5.12 ± 0.03 mg GAE/g), respectively ($p < 0.05$) (Table 1). Oregano extracts prepared under different process conditions, like solvent concentration and extraction time, showed values ranging from 5.76 to 18.75 mg GAE/g [37], while a higher level (39.48 mg GAE/g) was determined for an ethanolic extract [36].

Previous studies on parsley extracts revealed total phenolic contents ranging from 1.15 to 12.49 mg GAE/g [18, 35, 38, 39], which are consistent with the values found in this study. For basil, lower phenolic contents were assessed by Siti Mahirah et al. (2018) [33], ranging from 2.61 to 3.84 mg GAE/g.

For carotenoids, the levels decreased in the following order: 12.95 ± 0.09 µg/g (oregano) > 11.76 ± 0.12 µg/g (parsley) > 6.18 ± 0.04 µg/g (basil) ($p < 0.05$) (Table 1). Dobričević et al. (2019) [38] evaluated 6 cultivars of parsley, showing carotenoid levels from 80 to 160 µg/g. Higher contents were also reported for basil (829 µg/g) and oregano (345 µg/g) extracts [40].

The values found in the literature may differ significantly from those obtained in this study. Such variations are expected, as the levels of compounds like carotenoids and other phytochemicals are highly influenced by genetic factors and environmental conditions. Additionally, it is well established that the quantitative results of bioactive compounds in plant material extracts can be affected by extraction conditions, such as temperature, solvent type, solvent ratio, extraction time, and method [33, 37, 38].

Herbal extracts comprise a variety of components with diverse chemical structures and properties. Therefore, methods based on different mechanisms were employed to provide complementary insights. The DPPH method relies on the scavenging of the DPPH radical by antioxidants or radical species, resulting in the decolorization of the DPPH solution. In contrast, FRAP measures the reduction of the Fe(III) complex to Fe(II) in the presence of reducing agents [41].

Phenolic compounds are the primary class of antioxidants found in plant materials [42] and are strongly correlated with the high antioxidant capacity of natural extracts [43]. Additionally, carotenoids act as quenchers of singlet molecular oxygen and scavengers of free radicals, which are key reactive species driving oxidative processes [44].

3.2 Cooking effects on the total carotenoids content of omelets

Raw samples presented a total carotenoid amount of 14.87 ± 0.09 μg/g (Table 2), which is similar to the value reported by Panaite et al. (2021) [4] for fresh egg yolk (15.38 μg/g). However, differences may occur since carotenoid levels are primarily affected by the diet of laying hens [4, 8].

The heating process resulted in a decrease in carotenoid content in the control samples ($p < 0.05$). Total carotenoid levels reduced by 32.35% (from 14.87 ± 0.09 to 10.06 ± 0.11 μg/g) after air frying, 23.64% (from 14.87 ± 0.09 to 11.36 ± 0.10 μg/g) after microwaving, and 4.78% (from 14.87 ± 0.09 to 14.16 ± 0.47 μg/g) after pan frying. However, the decrease observed after pan frying was not statistically significant ($p > 0.05$) (Table 2).

The effects of cooking and high temperatures on carotenoid degradation are well documented [11, 12, 45]. Carotenoids are sensitive to heat, light, and oxygen, and can degrade during cooking due to oxidation and isomerization. These processes affect the functionality, bioavailability, and content of carotenoids [11, 12].

Studies have reported reduced levels of carotenoids, such as xanthophylls, in cooked eggs, which directly impacts their total carotenoid content. For example, the xanthophyll content in boiled, pan-fried, and microwaved egg yolk showed losses ranging from 6% to 18% [45]. Boiling and pan frying significantly decreased lutein and zeaxanthin levels in eggs, with reductions of approximately 13% and 24%, respectively, in fried samples [11].

After heating, the air-fried control samples exhibited the lowest carotenoid content, followed by the microwaved samples ($p > 0.05$). This suggests that carotenoid degradation during cooking is linked to both the duration of the cooking process and the temperature reached. As mentioned previously, omelets were air fried for 10 minutes, while microwaving and pan frying took 3 and 4 minutes, respectively. The cooking time and temperature for each method were determined through informal tests, taking sensory characteristics into account. Consequently, a longer cooking time was required to achieve the sensory characteristics deemed acceptable for air-fried omelets.

Although the time spent on pan frying was longer than microwaving, the internal temperature measured in omelets was higher in microwaved samples. The core temperatures in the samples decreased in the following order: air frying (138 °C) > microwaving (95 °C) > pan frying (84 °C). Pan-fried samples exhibited the lowest internal temperature, and the carotenoid content was similar to that of the raw samples. This behavior aligns with a thermal degradation kinetics study, which applied varying temperatures (25 °C, 35 °C, and 45 °C) and time intervals (0 - 10 hours) and showed that carotenoid contents decreased as heating time and temperature increased [12].

It is also important to note that during air frying hot air circulates inside the fryer chamber, and samples are most exposed to oxygen, a potent oxidation inducer, compared to the other cooking methods [15, 46]. Therefore, the continuous contact of food with the circulating air in the air fryer chamber is a critical factor.

Regarding omelet samples containing the herbs, they presented higher concentrations of carotenoids compared to control ($p < 0.05$) (Table 2). In air-fried samples added with herbs, the carotenoid amount ranged from 13.06 ± 0.10 to 14.63 ± 0.04 μg/g (basil), from 15.12 ± 0.05 to 18.00 ± 0.11 μg/g (oregano), and from 14.48 ± 0.64 to 16.41 ± 0.35 μg/g (parsley). For oregano differences were observed when comparing the different concentration of herb used (0.25, 0.5, or 0.75) ($p < 0.05$); however, for basil and parsley it was not noted for all levels. Regarding the addition of 0.75% herb, samples with oregano showed the highest carotenoid level (18.00 ± 0.11 μg/g), followed by parsley (16.41 ± 0.35 μg/g) and basil (14.63 ± 0.04 μg/g), respectively.

Table 2: Total carotenoids content (μg/g) of raw and cooked omelets (control and samples added with basil, oregano, and parsley at 0.25, 0.5, and 0.75 %).

	Air frying										
	Raw	**Control**	**Basil 0.25%**	**Basil 0.5%**	**Basil 0.75%**	**Oregano 0.25%**	**Oregano 0.5%**	**Oregano 0.75%**	**Parsley 0.25%**	**Parsley 0.5%**	**Parsley 0.75%**
Total Carotenoids (μg/g)	14.87 ± 0.09	$^{*;k}$10.06 ± 0.11$^{C;c}$	$^{*;h}$13.06 ± 0.10$^{B;e}$	$^{*;h}$13.24 ± 0.23$^{B;f}$	^{g}14.63 ± 0.04$^{A;f}$	^{f}15.12 ± 0.05$^{C;cd}$	$^{*;d}$17.00 ± 0.26 $^{B;cd}$	$^{*;c}$18.00 ± 0.11$^{A;bc}$	^{g}14.48 ± 0.64$^{B;d}$	$^{*;e}$16.35 ± 0.32$^{A;d}$	$^{*;e}$16.41 ± 0.35$^{A;d}$
	Pan frying										
	Raw	**Control**	**Basil 0.25%**	**Basil 0.5%**	**Basil 0.75%**	**Oregano 0.25%**	**Oregano 0.5%**	**Oregano 0.75%**	**Parsley 0.25%**	**Parsley 0.5%**	**Parsley 0.75%**
Total carotenoids (μg/g)	14.87 ± 0.09	^{g}14.16 ± 0.47$^{B;a}$	^{f}14.90 ± 0.08$^{AB;cd}$	^{f}15.17 ± 0.26$^{AB;e}$	$^{*;e}$15.88 ± 0.72$^{A;de}$	$^{*;d}$17.45 ± 0.16$^{A;ab}$	$^{*;c}$18.09 ± 0.67$^{A;ab}$	$^{*;b}$18.46 ± 0.62$^{A;ab}$	^{f}15.54 ± 0.06$^{C;c}$	$^{*;e}$16.38 ± 0.13$^{B;d}$	$^{*;d}$17.43 ± 0.20$^{A;\ c}$
	Microwaving										
	Raw	**Control**	**Basil 0.25%**	**Basil 0.5%**	**Basil 0.75%**	**Oregano 0.25%**	**Oregano 0.5%**	**Oregano 0.75%**	**Parsley 0.25%**	**Parsley 0.5%**	**Parsley 0.75%**
Total carotenoids (μg/g)	14.87 ± 0.09	$^{*;j}$11.36 ± 0.10 $^{D;b}$	$^{*;i}$12.11 ± 0.08$^{C;f}$	$^{*;g}$14.02 ± 0.11$^{B;f}$	^{f}15.13 ± 0.12$^{A;ef}$	$^{*;c}$17.98 ± 0.20$^{B;a}$	$^{*;c}$18.22 ± 0.10$^{B;a}$	$^{*;a}$19.24 ± 0.03$^{A;a}$	$^{*;d}$17.14 ± 0.40$^{B;b}$	$^{*;d}$17.37 ± 0.09$^{B;bc}$	$^{*;a}$19.41 ± 0.05$^{A;a}$

Results presented as mean ± standard deviation, n=3. "*" indicates significant differences in comparison with "Raw" by the Dunnet test. Similar lower letters on the left indicate that the treatments presented similar values by the Tukey test. Different capital letters on the right indicate differences among samples added with different concentrations of the same herb. Different lowercase letters on the right indicate differences among samples treated with different herbs at the same level of addition. All statistical analyses were performed at 5% of significance.

For pan frying, the highest content of carotenoids was found in samples added with 0.75% of oregano (18.46 ± 0.62 μg/g), followed by 0.75% of parsley (17.43 ± 0.20 μg/g), and 0.75% of basil (15.88 ± 0.72 μg/g). Using different levels of basil and oregano did not influence the results in samples containing these herbs ($p < 0.05$), which was not observed for parsley. In microwaved samples, the addition of basil, oregano, and parsley at 0.75% resulted in levels of 15.13 ± 0.12, 19.24 ± 0.03, and 19.41 ± 0.05 μg/g, respectively. The highest variation was determined for basil, with carotenoids varying from 12.11 ± 0.08 μg/g (0.25%) to 15.13 ± 0.12 μg/g (0.75%).

When comparing treatments with different herbs, samples containing oregano presented the highest carotenoid content ($p < 0.05$): 18.00 ± 0.11 μg/g (air frying), 18.46 ± 0.62 μg/g (pan frying), and 19.24 ± 0.03 μg/g (microwaving). However, an exception was noted for microwaved samples, which showed similar concentrations when oregano and parsley (0.75%) were used (19.41 ± 0.05 μg/g) ($p > 0.05$). These findings agree with the values presented in Table 1, where the highest content of carotenoids was assessed for oregano extract (12.95 ± 0.09 μg/g), followed by parsley (11.76 ± 0.12 μg/g). However, as indicated by the results for raw and control samples, heating led to carotenoid degradation in eggs, which may also occur with the carotenoids in the herbs.

It is well established that high cooking temperatures can impact food components in various ways. In the present study, air frying and microwaving significantly decreased the content of carotenoids ($p < 0.05$). Although pan cooking remains the most traditional method for cooking eggs, the current fast pace of life has influenced the way people prepare their foods, which reflects in the preference for more modern and convenient devices.

Air fryers have become increasingly popular as a healthier alternative for frying foods, as they eliminate the need for oil, thereby lowering the health risks linked to high-fat diets. However, the results of this study revealed that air frying had the most significant impact on carotenoid degradation. More extreme processing conditions were required to cook eggs during air frying, leading to greater degradation of carotenoids. Furthermore, both microwaving and air frying are considered cleaner, more practical, compact, and easier-to-use devices. They also offer precise process control and do not require gas for cooking.

Adding herbs to omelets resulted in higher levels of carotenoids (Figure 1). This can be attributed to the carotenoid content in the herbs (Table 1), which contributed additional amounts of these bioactive compounds to the omelets. The thermal stability of carotenoids depends on their chemical structures (e.g., the position and number of unsaturated double bonds, and hydroxyl groups) [12]. Additionally, the carotenoid profile in eggs differs from that of each herb studied. As a result, carotenoids may degrade differently, following distinct mechanisms. Furthermore, different cooking methods and conditions must be considered. It is important to note that similar behaviors regarding the degradation of carotenoids in eggs and herbs cannot be assumed, and further studies are needed to elucidate the mechanisms involved in the thermal degradation of carotenoids from both herbs and omelets separately.

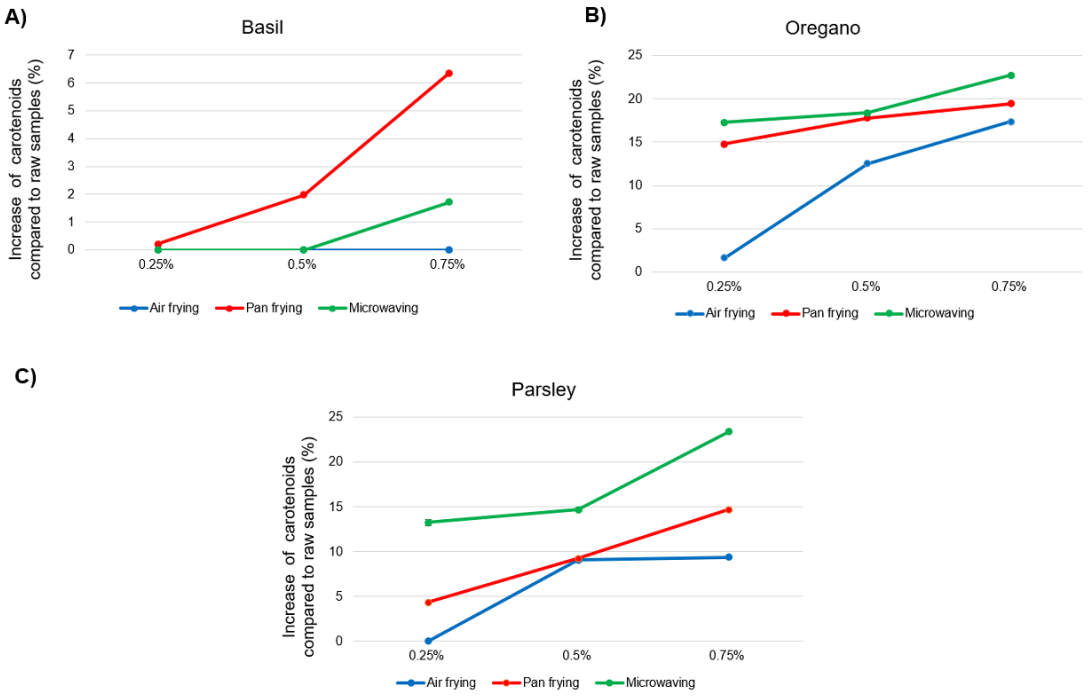

Figure 1: Percentage increase of carotenoid levels compared to raw samples for samples added with A) basil, B) oregano, and C) parsley.

Besides carotenoids, basil, oregano, and parsley contain other antioxidant compounds, such as phenolic compounds (Table 1), which may help protect the carotenoids in eggs from degradation processes like oxidation. The antioxidant potential of phenolic compounds is attributed to their ability to donate a hydrogen atom and/or an electron to free radicals, thereby preventing oxidative reactions. Additionally, phenolic compounds can protect molecules from initiators such as oxygen and metal ions [17, 20]. Since carotenoid oxidation can be triggered by factors such as light, heat, acidity, oxygen, metals, or interactions with radical species [47], phenolic compounds from herbs may help minimize carotenoid losses in eggs during heating.

4. CONCLUSION

The overall results highlight the significant degradative effects of air frying and microwaving on the total carotenoid content in omelets, with air-fried control samples showing the lowest levels of carotenoids. However, samples containing herbs exhibited higher carotenoid concentrations, which may be attributed to the additional carotenoids from the herbs and their protective effect against carotenoid degradation in eggs. Among the herbs, the most promising results were determined for oregano, mainly when it was applied at 0.75%. Therefore, adding herbs to omelets can be considered a suitable domestic strategy, not only to enhance organoleptic qualities but also to minimize thermal degradation of carotenoids in eggs and add extra nutritional value. Moreover, different mechanisms may be involved in the degradation of carotenoids from various sources (eggs and herbs) during different cooking techniques, suggesting the need for further studies to better elucidate these findings.

5. ACKNOWLEDGMENT

The authors thank the Natl. Brazilian Research Foundations (CAPES) for the scholarship and FAPERJ (Foundation for Research of the Rio de Janeiro State).

6. REFERENCES

1. Réhault-Godbert S, Guyot N, Nys Y. The golden egg: nutritional value, bioactivities, and emerging benefits for human health. Nutrients. 2019 Mar;11(3):684. doi: 10.3390/nu11030684
2. Sass CAB, Pimentel TC, Aleixo MGB, Dantas TM, Cyrino Oliveira FL, de Freitas MQ, et al. Exploring social media data to understand consumers' perception of eggs: A multilingual study using Twitter. J Sens Stud. 2020 Sep;35(6):e12607. doi: 10.1111/joss.12607
3. Razi SM, Fahim H, Amirabadi S, Rashidinejad A. An overview of the functional properties of egg white proteins and their application in the food industry. Food Hydrocoll. 2023 Feb;135:108183. doi: 10.1016/j.foodhyd.2022.108183
4. Panaite TD, Nour V, Saracila M, Turcu RP, Untea AE, Vlaicu PA. Effects of linseed meal and carotenoids from different sources on egg characteristics, yolk fatty acid and carotenoid profile and lipid peroxidation. Foods. 2021 May;10(6):1246. doi: 10.3390/foods10061246
5. Lu Y, Hao YJ, Zhou X, Huang F, Li C, Wang J, et al. Effects of long-term intake of carotenoid-enriched eggs on healthy people: a randomized controlled study. Food Funct. 2024 Jun;15:7032-45. doi: 10.1039/D4FO00910J
6. Crupi P, Faienza MF, Naeem MY, Corbo F, Clodoveo ML, Muraglia M. Overview of the potential beneficial effects of carotenoids on consumer health and well-being. Antioxidants. 2023 May;12(5):1069. doi: 10.3390/antiox12051069
7. Alvarado K, Bordat C, Reboul E. Egg carotenoids and eye health. In: Mine Y, Guyonnet V, Hatta H, Nau F, Qiu N, editors. Handbook of egg science and technology. Boca Raton (US): CRC Press; 2023. p. 681-704.
8. Kljak K, Carović-Stanko K, Kos I, Janječić Z, Kiš G, Duvnjak M, et al. Plant carotenoids as pigment sources in laying hen diets: Effect on yolk color, carotenoid content, oxidative stability and sensory properties of eggs. Foods. 2021 Mar;10(4):721. doi: 10.3390/foods10040721
9. Black HS, Boehm F, Edge R, Truscott TG. The benefits and risks of certain dietary carotenoids that exhibit both anti-and pro-oxidative mechanisms—A comprehensive review. Antioxidants. 2020 Mar;9(3):264. doi: 10.3390/antiox9030264
10. Maliar T, Maliarová M, Blažková M, Kunštek M, Uváčková Ľ, Viskupičová J, et al. Simultaneously determined antioxidant and pro-oxidant activity of randomly selected plant secondary metabolites and plant extracts. Molecules. 2023 Sep;28(19):6890. doi: 10.3390/molecules28196890
11. Nimalaratne C, Schieber A, Wu J. Effects of storage and cooking on the antioxidant capacity of laying hen eggs. Food Chem. 2016 Mar;194:111-6. doi: 10.1016/j.foodchem.2015.07.116
12. Xiao YD, Huang WY, Li DJ, Song JF, Liu CQ, Wei QY, et al. Thermal degradation kinetics of all-trans and cis-carotenoids in a light-induced model system. Food Chem. 2018 Jan;239:360-8. doi: 10.1016/j.foodchem.2017.06.107
13. Kelly AL, vo Kientza HT. Microwave heating and modern cuisine. In: vo Kientza HT, Lavelle C, Burke R, Kelly A L, editors. Handbook of molecular gastronomy. Boca Raton (US): CRC Press; 2021. p. 429-32.
14. de Oliveira VS, Viana DSB, Keller LM, de Melo MTT, Mulandeza OF, Barbosa MIMJ, et al. Impact of air frying on food lipids: Oxidative evidence, current research, and insights into domestic mitigation by natural antioxidants. Trends Food Sci Technol. 2024 May;147:104465. doi: 10.1016/j.tifs.2024.104465
15. Ferreira FS, Sampaio GR, Keller LM, Sawaya AC, Chávez DW, Torres EA, et al. Impact of air frying on cholesterol and fatty acids oxidation in sardines: Protective effects of aromatic herbs. J Food Sci. 2017 Nov;82(12):2823-31. doi: 10.1111/1750-3841.13967
16. De Pilli T, Alessandrino O. Effects of different cooking technologies on biopolymers modifications of cereal-based foods: Impact on nutritional and quality characteristics review. Crit Rev Food Sci Nutr. 2020 Dec;60(4):556-65. doi: 10.1080/10408398.2018.1544884
17. Embuscado ME. Bioactives from culinary spices and herbs: a review. J Food Bioact. 2019;6:68-99. doi: 10.31665/JFB.2019.6186
18. Ferreira FS, de Oliveira VS, Chávez DWH, Chaves DS, Riger CJ, Sawaya ACHF, et al. Bioactive compounds of parsley (*Petroselinum crispum*), chives (*Allium schoenoprasum* L) and their mixture (Brazilian cheiro-verde) as promising antioxidant and anti-cholesterol oxidation agents in a food system. Food Res Int. 2022 Jan;151:110864. doi: 10.1016/j.foodres.2021.110864
19. Kulawik A, Cielecka-Piontek J, Zalewski P. The importance of antioxidant activity for the health-promoting effect of lycopene. Nutrients. 2023 Aug;15(17):3821. doi: 10.3390/nu15173821
20. Zeb A. Phenolic antioxidants in foods: Chemistry, biochemistry and analysis. Berlin/Heidelberg (DE): Springer; 2021.

21. de Oliveira VS, Chávez DWH, Paiva PRF, Gamallo OD, Castro RN, Sawaya ACHF, et al. Parsley (*Petroselinum crispum* Mill.): A source of bioactive compounds as a domestic strategy to minimize cholesterol oxidation during the thermal preparation of omelets. Food Res Int. 2022 Jun;156:111199. doi: 10.1016/j.foodres.2022.111199
22. Mariano BJ, de Oliveira VS, Chávez DWH, Castro RN, Riger CJ, Mendes JS, et al. Biquinho pepper (*Capsium chinense*): Bioactive compounds, in vivo and in vitro antioxidant capacities and anti-cholesterol oxidation kinetics in fish balls during frozen storage. Food Biosci. 2022 Jun;47:101647. doi: 10.1016/j.fbio.2022.101647
23. Swain T, Hillis WE. The phenolic constituents of *Prunus domestica*. I.—The quantitative analysis of phenolic constituents. J Sci Food Agric. 1959 Feb;10(1):63-8. doi: 10.1002/jsfa.2740100211
24. Rufino MSM, Alves RE, de Brito ES, Pérez-Jiménez J, Saura-Calixto F, Mancini-Filho J. Bioactive compounds and antioxidant capacities of 18 non-traditional tropical fruits from Brazil. Food Chem. 2020 Aug;121(4):996-1002. doi: 10.1016/j.foodchem.2010.01.037
25. Thaipong K, Boonprakob U, Crosby K, Cisneros-Zevallos L, Byrne DH. Comparison of ABTS, DPPH, FRAP, and ORAC assays for estimating antioxidant activity from guava fruit extracts. J Food Compos Anal. 2006 Sep;19(6-7):669-75. doi: 10.1016/j.jfca.2006.01.003
26. Lichtenthaler HK. Chlorophylls and carotenoids: pigments of photosynthetic biomembranes. In: Parker L, Douce R, editors. Methods in enzymology. New York: Academic Press; 1987. p. 350-82.
27. de Oliveira VS, Chávez DWH, Gamallo OD, Castro RN, Sawaya ACHF, Sampaio GR, et al. Effect of bioactive compounds from culinary herbs against cholesterol thermo-oxidation in omelets during home cooking processes. Int J Gastron Food Sci. 2024 Nov;38:101077. doi: 10.1016/j.ijgfs.2024.101077
28. de Oliveira VS, Chávez DWH, Paiva PRF, Gamallo OD, Castro RN, Sawaya ACHF, et al. Parsley (*Petroselinum crispum* Mill.): A source of bioactive compounds as a domestic strategy to minimize cholesterol oxidation during the thermal preparation of omelets. Food Res Int. 2022 Jun;156:111199. doi: 10.1016/j.foodres.2022.111199
29. Brazilian Health Regulatory Agency [Agência Nacional de Vigilância Sanitária] (ANVISA). Resolução – RDC nº 216, de 15 de setembro de 2004. Dispões sobre o regulamento técnico de boas práticas de serviço de alimentação. Brasília (DF): Diário Oficial da União; 2024. Available from: https://bvsms.saude.gov.br/bvs/saudelegis/anvisa/2004/res0216_15_09_2004.html
30. Rodriguez-Amaya DB. A guide to carotenoid analysis in foods. Washington, DC (US): ILSI press; 2001.
31. Nikolic JS, Mitic VD, Stankov Jovanovic VP, Dimitrijevic MV, Stojanovic GS. Chemometric characterization of twenty-three culinary herbs and spices according to antioxidant activity. J Food Meas Charact. 2019 Apr;13:2167-76. doi: 10.1007/s11694-019-00137-0
32. Jadczak D, Bojko K, Wysocka G, Szymanska M. Yield and biological properties of leaf parsley (*Petroselinum crispum* (Mill.) Nym. Ex AW Hillc Convar. crispum). J Elem. 2019;24(2):804-815. doi: 10.5601/jelem.2018.23.4.1697
33. Siti Mahirah Y, Rabeta MS, Antora RA. Effects of different drying methods on the proximate composition and antioxidant activities of *Ocimum basilicum* leaves. Food Res. 2018 Oct;2(5):421-8. doi: 10.26656/fr.2017.2(5).083
34. Hossain MB, Patras A, Barry-Ryan C, Martin-Diana AB, Brunton NP. Application of principal component and hierarchical cluster analysis to classify different spices based on in vitro antioxidant activity and individual polyphenolic antioxidant compounds. J Funct Foods. 2011 Jul;3(3):179-89. doi: 10.1016/j.jff.2011.03.010
35. Mazzucotelli CA, González-Aguilar GA, Villegas-Ochoa MA, Domínguez-Avila AJ, Ansorena MR, Di Scala KC. Chemical characterization and functional properties of selected leafy vegetables for innovative mixed salads. J Food Biochem. 2018 Nov;42(1):e12461. doi: 10.1111/jfbc.12461
36. Parra C, Muñoz P, Bustos L, Parra F, Simirgiotis MJ, Escobar H. UHPLC-DAD characterization of *Origanum vulgare* L. from Atacama desert Andean region and antioxidant, antibacterial and enzyme inhibition activities. Molecules. 2021 Apr;26(7):2100. doi: 10.3390/molecules26072100
37. Majeed M, Hussain AI, Chatha SA, Khosa MK, Kamal GM, Kamal MA, et al. Optimization protocol for the extraction of antioxidant components from *Origanum vulgare* leaves using response surface methodology. Saudi J Biol Sci. 2016;23(3):389-96. doi: 10.1016/j.sjbs.2015.04.010
38. Dobričević N, Šic Žlabur J, Voća S, Pliestić S, Galić A, Delić A, et al. Bioactive compounds content and nutritional potential of different parsley parts (*Petroselinum crispum* Mill.). J Cent Eur Agric. 2019;20(3):900-10. doi: 10.5513/JCEA01/20.3.2417
39. Epifanio NM, Cavalcanti LRI, dos Santos KF, Duarte PSC, Kachlicki P, Ożarowski M, et al. Chemical characterization and *in vivo* antioxidant activity of parsley (*Petroselinum crispum*) aqueous extract. Food Funct. 2020 Apr;11(6):5346-56. doi: 10.1039/D0FO00484G

40. Stancheva I, Geneva M, Markovska Y, Tzvetkova N, Mitova I, Todorova M, et al. A comparative study on plant morphology, gas exchange parameters, and antioxidant response of *Ocimum basilicum* L. and *Origanum vulgare* L. grown on industrially polluted soil. Turk J Biol. 2014;38(1):89-102. doi: 10.3906/biy-1304-94
41. Gulcin İ. Antioxidants and antioxidant methods: An updated overview. Arch Toxicol. 2020 Mar;94(3):651-715. doi: 10.1007/s00204-020-02689-3
42. Nardini M. Phenolic compounds in food: Characterization and health benefits. Molecules. 2022 Jan;27(3):783. doi: 10.3390/molecules27030783
43. Butkevičiūtė A, Urbštaitė R, Liaudanskas M, Obelevičius K, Janulis V. Phenolic content and antioxidant activity in fruit of the genus *Rosa L*. Antioxidants. 2022 May;11(5):912. doi: 10.3390/antiox11050912
44. Young AJ, Lowe GL. Carotenoids—antioxidant properties. Antioxidants. 2018 Feb;7(2):28. doi: 10.3390/antiox7020028
45. Nimalaratne C, Lopes-Lutz D, Schiebe A, Wu J. Effect of domestic cooking methods on egg yolk xanthophylls. J Agric Food Chem. 2012 Dec;60(51):12547-52. doi: 10.1021/jf303828n
46. de Carvalho IOAM, de Oliveira VS, Chávez DWH, Gamallo OD, Castro RN, Sawaya ACHF, et al. The use of lemon juice and its role on polyunsaturated fatty acids and cholesterol oxides formation in thermally prepared sardines. J Food Compos Anal. 2021 Dec;104:104087. doi: 10.1016/j.jfca.2021.104087
47. Butnariu M. Methods of analysis (extraction, separation, identification and quantification) of carotenoids from natural products. J Ecosyst Ecography. 2016;6(2):1-19. doi: 10.4172/2157-7625.1000193