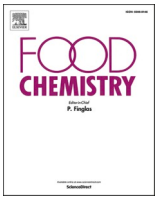
Food Chemistry 442 (2024) 138368

Contents lists available at ScienceDirect

Food Chemistry

journal homepage: www.elsevier.com/locate/foodchem



# Impact of washing and freezing on nutritional composition, bioactive compounds, antioxidant activity and microstructure of mango peels

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ARTICLE INFO

*Keywords:*
Mango byproducts
Washing
Freezing
vitamin C
Phenolic compounds
Carotenoids

ABSTRACT

Mango peels are widely produced and highly perishable. Disinfectant washing and freezing are among the most used methods to preserve foods. However, their impact on products' properties is conditioned by the foods' features. This study evaluated for the first time the phytochemical composition, antioxidant activity, and microstructure of mango peels washed with peracetic acid (27 mg/mL for 19 min) and frozen at −20 °C for 30 days. Washing decreased the content of vitamin C (−7%), penta-O-galloyl-β-D-glucose (−23 %), catechin (−30 %), and lutein (−24 %), but the antioxidant activity was preserved. Freezing changed mango peels' microstructure, increased free phenolic compounds, namely acid gallic (+36 %) and catechin (+51 %), but reduced bound phenolic compounds (−12 % to −87 %), bound phenolic compounds' antioxidant activity (−51 % to −72 %), and violaxanthin (−51 %). Both methods were considered adequate to conserve mango peels since fiber and the main bioactive compounds (free mangiferin, free gallic acid, and β-carotene) remained unchanged or increased.

## 1. Introduction

Mango (*Mangifera indica* L.) is a climacteric fruit produced in regions with tropical or subtropical climates but worldwide appreciated and consumed (Brecht & Sidhu, 2017). In 2021, the world production of mangoes, mangosteens, and guavas was 62.84 million tons (Shahbandeh, 2023). Mangoes represented the most of this quantity (about 75 %) (Brecht & Sidhu, 2017). Usually, they are peeled before being consumed as a dessert fruit or processed (Kaur et al., 2023), so as peels constitute 15–20 % of mangoes' total weight (Nguyen et al., 2019), a huge amount of mango peels is generated as a byproduct. Hence, in 2021, it can be estimated that 7–9 million tons of mango peels were produced worldwide, but most of them were discarded in landfills or incinerated, contributing to the severe environmental problems that humanity currently faces (Nguyen et al., 2019; Shahbandeh, 2023). Nevertheless, mango peels are a great source of fiber and antioxidants, namely, xanthones, flavonoids, gallic acid derivates, vitamin C, and carotenoids, and, therefore, they have a high potential to be upcycled in the food industry (Kaur et al., 2023; Marçal & Pintado, 2021).

Like other fruit byproducts, mango peels are a highly perishable raw material. Besides they have a high water content, during the peeling process, cells are ruptured, which accelerates browning, respiration rate, softening, and microbial growth and, consequently, impairs their food safety, nutritional value, bioactivity, and sensory properties (De Corato, 2020; Marçal & Pintado, 2021). So, mango peel processing into functional ingredients usually starts with a disinfectant washing to decrease the organic and microbial load. Marçal et al. (2022) evaluated for the first time the effect of washing conditions (food product to disinfectant solution ratio, sodium hypochlorite (SH) or peracetic acid (PAA) concentration, and disinfection time) on mango peels' microbial load, phytochemical composition (dry matter, total phenolic compounds, and total carotenoids) and antioxidant activity. Succinctly, results showed that high disinfectant concentrations and disinfection times positively impacted microbiological quality but decreased total carotenoids and antioxidant activity. The same study estimated optimal washing conditions with SH or PAA to simultaneously maximize the reduction of microbial load and the preservation of phytochemical composition and antioxidant activity (Marçal et al., 2022).

After disinfectant washing, other processing methods should be applied to preserve and process mango peels into functional ingredients

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https://doi.org/10.1016/j.foodchem.2024.138368
Received 21 August 2023; Received in revised form 1 January 2024; Accepted 3 January 2024
Available online 8 January 2024

or extract from them natural additives. Drying has been pointed out as the best option to process mango peels since it enables preserving them and, when combined with milling, facilitates their incorporation into several foods (Marçal et al., 2022). However, depending on applied conditions, drying can damage important mango peels' bioactive compounds, namely xanthones, flavonoids, and alk(en)ylresorcinols (Ancos et al., 2018; Geerkens et al., 2015). Although this option has been scarcely explored, freezing is possibly a viable alternative to preserving mango peels. Incorporating fresh/frozen mango peels in food products can be facilitated by developing pulps. Additionally, freezing can be used as a complementary preserving method (when processing industries cannot dry mango peels during their shelf-life) or pretreatment to reduce drying time (Marçal & Pintado, 2021; Noshad & Ghasemi, 2020).

The effect of freezing on the characteristics of edible fruits has been extensively studied (Bonat Celli et al., 2016). However, the published data are often quite discrepant. For instance, considering the impact of freezing on fruits' phenolic compounds, the results varied between significant increases, no effect, and marked decreases, even when similar processing and storage conditions were applied (Araújo-Rodrigues et al., 2021; Bonat Celli et al., 2016). Regarding the application of freezing as a pretreatment before drying, there is still little information available. Noshad and Ghasemi (2020) showed that prior freezing reduced grapes' drying time and improved total phenolic compounds and antioxidant activity preservation. On the other hand, Stamenković et al. (2019) reported that freezing before drying at 60, 70, and 80 °C and air velocity of 1.5 m/s decreased dried raspberries' total phenolic compounds. Therefore, the freezing effect on vegetal foods' characteristics, whether applied as a preservation method or as a pretreatment, is conditioned not only by processing and storage conditions but also by the intrinsic features of foods.

A search on the Web of Science was performed to find all papers with the words "mango" and "peels" in their titles, abstracts, or keywords. After analyzing all of them, it was concluded that at this moment, there are no studies about the impact of freezing on mango peels' phytochemical composition, bioactivity, and microstructure. Furthermore, the effect of disinfectant washing with PAA on mango peels' soluble and insoluble fiber content, vitamin C, phenolic compounds profile, and carotenoids profile remained unknown. As mentioned before, disinfection and preservation methods are essential processing steps to preserve mango peels' food safety, nutritional quality, and bioactivity and, consequently, to enable the valorization of this byproduct in the food and pharmaceutical industries. Washing with PAA is among the safest and cheapest procedures to disinfect fruits and vegetables, while freezing is one of the most used methods to preserve foods (Bonat Celli et al., 2016; Marçal et al., 2022). However, considering the lack of knowledge described above, identifying the main advantages and disadvantages of these two processing methods within mango peel phytochemical preservation, it is essential to provide useful information for researchers, processing companies, and other stakeholders to choose these or other alternative methods considering the desired characteristics of the product that they intend to develop. Hence, the present study aimed to evaluate the impact of washing with PAA and freezing on mango peels' nutritional composition, free and bound phenolic compounds profiles, carotenoids profile, and antioxidant activity. Furthermore, the freezing effect on the microstructure of mango peels was assessed through scanning electron microscopy (SEM).

## 2. Materials and methods

### 2.1. Chemicals

The commercial disinfectant with PAA (Mida Chriox 5) was purchased from Christeyns (Agualva - Cacém, Portugal). Acetone, dichloromethane and glutaraldehyde were provided by Honeywell Fluka (North Carolina, USA). Hexane and absolute ethanol were purchased from Carlos Erba Reagents (Barcelona, Spain). Methanol was acquired at VWR Chemicals (Pennsylvania, USA). Ethyl acetate and acetonitrile were provided by Fisher Chemical (New Hampshire, USA). Trifluoroacetic acid, phosphate buffer saline (PBS), ascorbic acid, dithiothreitol, phosphoric acid and KH2PO4 were purchased from Sigma-Aldrich (St. Louis, USA). All phenolic compounds and carotenoids standers were purchased from Sigma - Aldrich (St. Louis, USA) or Extrasynthese (Genay, France). The reagents used to evaluate antioxidant activity namely, 2-azinobis-3-ethylbenzothiazoline-6-sulphonic acid (ABTS), 2,2-azo-bis-(2-methylpropionamidine)-dihydrochloride (AAPH), 2,2-diphenyl-1-picrylhydrazyl (DPPH), Folin-Ciocalteu's reagent and Trolox were purchased from Sigma-Aldrich (St. Louis, USA).

### 2.2. Mango peels processing

Mango peels used to perform all analyses, excluding microstructure, were provided by a Portuguese company (Nuvi Fruits, SA) that produces minimally processed fruits. They were obtained from Mangifera indica L. 'Tommy Atkins,' harvested at the mature green stage, transported from Brazil to Portugal (at 8–10 °C and 80 % of relative humidity for 8 days), and then ripened post-harvested (at room temperature, up to ≥12 °bx).

Firstly, mango peels were cut into pieces of about 5 g and mixed to improve sample homogeneity. After they were randomly divided into 9 equal parts: 3 parts were not submitted to any treatment **(unprocessed samples (UNP));** three parts were washed with PAA according to the optimal conditions (mango peels to disinfectant solution ratio: 1:1 (kg:L); PAA concentration: 27 mg/mL; disinfection time: 19 min) determined by Marçal et al. (2022) **(washed samples (W))**; and the other three parts were also washed as described and, then frozen at −20 °C (Whirlpool WHE31352FO2) for 30 days **(frozen samples (FZ)).**

Peels used to perform the microstructure analyses were also from Tommy Atkins mangoes cultivated in Brazil but purchased in a supermarket. First, the mangoes were washed with tap water and peeled to obtain byproducts (peels) like those supplied by Nuvi Fruits. Afterward, the peels were washed with PAA as aforementioned and kept at 6 °C (W) or −20 °C (FZ) for 24 h.

### 2.3. Samples preparation for nutritional composition analysis and bioactive compounds extraction

Fresh and frozen mango peels were used to determine moisture, protein, and ash and extract vitamin C, phenolic compounds, and carotenoids. Immediately before performing these analyses, to ensure the samples' representativeness, about 50 g of mango peels were milled using a grinder (Coffee Grinder TAURUS Aromatic II, Barcelona, Spain). On the other hand, other composition parameters such as total fat, soluble and insoluble fiber were determined from dried mango peels. Fresh and frozen samples were hot air dried at 65 °C for 48 h, with constant air circulation (air flow: 1.6 m/s; relative humidity: 53 %) and milled with the grinder.

### 2.4. Macronutrients and ash

Moisture, protein (factor conversion: 6.25), and ash were determined according to the AOAC standards (1990) methods. The procedure described by Gómez-García et al. (2021) was carried out to determine the total fat. Carbohydrates were estimated by difference [100 − (% moisture + % ash + % protein + % lipid)]. Finally, soluble and insoluble dietary fiber were analyzed following the procedure presented in Megazyme (2017). All results were expressed in g/100 g of fresh weight (FW).

### 2.5. Vitamin C

#### 2.5.1. Extraction

The method described by Brause et al. (2003) was followed to extract and quantify vitamin C. Briefly, samples (4 g) were homogenized with 15 mL of ultra-pure water using a dispersing machine ((IKA Ultra-turrax T18, Wilmington, USA) at 19 000 rpm for 1 min. After, the supernatants were separated from the residue by centrifugation (3 900 *g*; 4 °C; 20 min) (centrifuge, UNIVERSAL R 320, Hettich, Tuttlingen, Germany). Then, ca. 1 mg of dithiothreitol was added per mL of extract recovered, and they were kept at room temperature and protected from light for 2 h. Finally, extracts were filtered with a 0.22 μm membrane and analyzed by HPLC (Brause et al., 2003).

#### 2.5.2. Quantification by HPLC

Vitamin C was quantified using an HPLC Agilent interfaced with a 1260 Infinity II Diode Array Detector WR. The stationary phase was a silica column (ACE equivalence 5 mm, C18, 250 × 4.6 mm column), while the mobile phase comprised $KH_2PO_4$ (0.5 %, w/v) and dithiothreitol (0.1 %, w/v) dissolved in ultra-pure water. The pH of the mobile phase was adjusted to 2.5 with phosphoric acid. The flow and injection volumes were 0.5 mL/min and 50 μl, respectively. The detection was carried out at 254 nm. All the process occurred at room temperature. Data acquisition and analysis were made using Software Data Analysis Agilent.

The calibration curve was performed with ascorbic acid (5–40 μg/mL; $R^2$ = 99.33 %) and results were presented in mg/100 g FW.

### 2.6. Phenolic compounds

#### 2.6.1. Methanolic extraction of free phenolic compounds

Free phenolic compounds were released from mango peels through a methanolic extraction following the procedure described in our previous work (Marçal et al., 2022). Succinctly, mango peels were homogenized with methanol (extraction conditions: 1 g of mango peels dry weight (DW) to 25 mL of methanol 80 %, considering the samples average DW) using a dispersing machine ((IKA Ultra-turrax T18, Wilmington, USA) at 24 000 rpm for 1 min. Then, the slurry was centrifugated (3 900 *g*, 20 min, 4 °C) to separate extracts from residues. The free phenolic compounds extracts **(FPC)** were kept at −80 °C until they were used to identify and quantify free phenolic compounds and evaluate antioxidant activity. Residues were stored at −20 °C until they were submitted to basic and acid hydrolyses to release bound phenolic compounds from the matrix.

#### 2.6.2. Basic and acid hydrolyses to determine bound phenolic compounds

The basic and acid hydrolyses were carried out according to Pacheco-Ordaz et al. (2018), with some modifications. Succinctly, 20 mL of NaOH 4 M was added to the residues obtained from FPC extracts. The slurries were agitated for 4 h at 250 rpm and room temperature using an Orbital Shaker MaxQ 6000 (Thermo Scientific, Waltham, USA). After, the pH was adjusted to 2.0 using HCl 6 M. Then, centrifugation (3 900 *g*, 20 min, 4 °C) was performed to separate the supernatants from the remaining residues. Finally, the supernatants were extracted three times with ethyl acetate.

The remaining residues from basic hydrolyses were submitted to acid hydrolysis with 5 mL of HCl 2 M at 85 °C for 1 h. Then, the same steps performed in the basic hydrolysis were carried out. The pH adjustment was done with NaOH 4 M.

The extracts (ethyl acetate) from basic and acid hydrolyses (**BBPC** and **ABPC**, respectively) were evaporated at 30 °C to dryness in an RVC 2–18 speed-vacuum evaporator (Christ, Osterode am Harz, Germany), resuspended in 2 mL of methanol 80 % and filtered with a 0.45 μm membrane.

#### 2.6.3. Total phenolic compounds

The total phenolic compounds were determined through the Folin–Ciocalteu method following the procedure performed by Vilas-Boas et al. (2020). The calibration curve was performed with gallic acid (0.01–0.10 mg/mL; $R^2$ = 99. 78 %). The results were expressed in μg of gallic acid equivalents (GAEs)/*g* FW.

#### 2.6.4. Identification and quantification by HPLC

The identification of main phenolic compounds was carried out in an HPLC Waters Alliance e2695 separation module system interfaced with a photodiode array UV/Vis detector 2998 (PDA 190–600 nm) (Waters, Milford, USA)) and according to Vilas-Boas et al. (2020). The separation occurred in a reversed column (ZORBAX Eclipse XDB-C18, 80 Å, 4.6 × 250 mm, 5 μm (Agilent, Santa Clara, USA)) at 25 °C. Two different mobile phases (A and B) were used. Mobile phase A comprised 94.9 % of ultrapure water, 5 % of acetonitrile, and 0.1 % of trifluoroacetic acid (v:v:v), while mobile phase B contained 99,9% of acetonitrile and 0.1 % of trifluoroacetic acid (v:v:v). The gradient elution applied was: 0–1 min 100 % A; 1–30 min 79 % A; 30–42 min 73 % A; 45–55 min 42 % A; 55–60 min 100 % A. The flow and injection volume were 1 mL/min and 20 μl, respectively.

Data acquisition and analysis were carried out using Software Empower 3. Detection was made at three different wavelengths (280 nm, 320 nm, and 350 nm). Identification and quantification of phenolic compounds were made by comparing peaks' retention time, UV absorption spectrum, and area with calibration curves of pure standards (mangiferin: 1.95–250 μg/mL; gallic acid: 1.95–500 μg/mL; penta-O-galloyl-β-D-glucose, methyl gallate and 4-hydroxybenzoic acid: 0.49–125 μg/mL; catechin: 0.98–250 μg/mL; quercetin-3-O-galactoside, quercetin 3-β-D-glucoside, quercetin 3-O-α-L-arabinopyranoside and quercetin-3-O-α-L-arabinofuranoside: 0.24–125 μg/mL; ferulic acid and p-coumaric acid: 0.49–62.50 μg/mL; 3,4-dihydroxybenzoic acid: 0.49–250 μg/mL; $R^2$: 99.99 % − 100 %). The results were shown in μg/g FW.

### 2.7. Carotenoids

#### 2.7.1. Extraction

Carotenoids extraction and saponification were performed based on the method used by Stinco et al. (2014) and exactly as described by Marçal et al. (2022). After saponification, extracts were washed with ultra-pure water, evaporated at 30 °C to dryness in an RVC 2–18 speed-vacuum evaporator (Christ, Osterode am Harz, Germany), and resuspended in 2 mL of hexane: acetone (1:1; v:v).

#### 2.7.2. Identification and quantification by HPLC

Carotenoids were analyzed using an HPLC Agilent interfaced with a 1260 Infinity II Diode Array Detector WR, and according to the method described by Oliveira et al. (2004), with slight modifications. The stationary phase was a reversed column (ZORBAX Eclipse XDB-C18, 80 Å, 4.6 × 250 mm, 5 μm (Agilent, Santa Clara, USA)). In its turn, two mobile phases were used: A (100 % ethyl acetate) and B (90 % of acetonitrile and 10 % of water (v:v)). The gradient employed was: 0–31 min (0–60 % A); 31–46 min (60 % A); 46–51 min (60–100 % A); 51–55 min (100 % A); 55–60 min (100—0 % A); 60–65 min (0 % A). The flow and injection volume were 0.8 mL/min and 20 μl, respectively. All the process occurred at room temperature.

The detection was carried out at 470 nm. Data acquisition and analysis were made using Software Data Analysis Agilent. Identification and quantification of carotenoids were carried out by comparing peaks' retention time, UV absorption spectrum, and area with calibration curves of pure standards (β-carotene: 1.81–100 μg/mL, $R^2$: 98.96 %; lutein: 0.93–7.26 μg/mL, $R^2$: 99.82 %; violaxanthin: 0.23–3–75 μg/mL, $R^2$: 99.47 %). The results were depicted in μg/g FW.

### 2.8. Antioxidant assays

FPC, BBPC, and ABPC extracts, described in Sections 2.6.1 and 2.6.2, were submitted to ABTS and DPPH assays. Both methods were carried out as described by Vilas-Boas et al. (2020), and their calibration curves were performed with Trolox (6–63 μg/ml; $R^2$: 98.28 %–99.70 %). The results were expressed in μg of Trolox equivalents (TEs)/g FW.

### 2.9. Microstructure

The microstructure of fresh and frozen samples was analyzed through SEM. The preparation of samples included three steps: fixation, dehydration, and drying. Fixation and dehydration were performed according to Almeida et al. (2022), with some modifications. Firstly, peels were vertically cut with a scalpel to their thickness <1 mm, and the analyzed piece included all constituents of samples, namely epicarp (peel) and mesocarp (pulp) (Fig. 1). Afterward, samples were immersed in a fixation solution composed of glutaraldehyde and PBS 0.1 M (2.5 %:97.5 %; v:v) for 90 h. Then, samples were rinsed with ultra-pure water four times and dehydrated by immersion in an ascending ethanol gradient (30–100 %) for 1 h in each solution. Samples were kept submerged in the ethanol 100 % overnight, and on the following day, they were dried in a Nova Sterilis Nova Genesis Supercritical $CO_2$ reactor at 40 °C, 100 bar, for 45 min. Finally, dried peels were placed on top of observation pins, coated with gold/palladium as described by Almeida et al. (2022), and their microstructure was observed using a Phenom XL G2 desktop scanning electron microscope (Thermo Fischer Scientific, The Netherlands), at an accelerating voltage of 5 kV. Observations were performed, and micrographs were collected using the secondary electron detector (SED).

### 2.10. Statistical analysis

The statistical analysis was done with IMB SPSS Statistics Software (New York, USA). *T*-student test for independent samples was applied to compare UNP × W and W × FZ samples and, hence, evaluate the effect of washing and freezing, respectively. Both treatments were performed in triplicate (Section 2.2).

## 3. Results

### 3.1. Macronutrients, ash, and vitamin C

Table 1 displays the nutritional composition of UNP, W, and FZ mango peels. They were mostly composed of water (>80 g/100 g FW) and carbohydrates (>15 g/100 g FW). Total fiber varied between 5.60 and 5.87 g/100 g FW, corresponding to more than 35 % of total carbohydrates. The soluble/insoluble fiber ratios were approximately 1. Regarding the other macronutrients, results showed that mango peels had a low amount of protein (0.72–0.99 g/100 g FW) and fat (0.26–0.34 g/100 g FW). Finally, ash and vitamin C content ranged between 0.35–0.42 g/100 g FW and 4.57–5.20 mg/100 g FW, respectively.

Washing caused a reduction of 7 % in vitamin C content. Furthermore, although no statistically significant differences were detected, compared with UNP, the W mango peels tended to have a lower dry weight, total carbohydrates, soluble fiber, and ash. On the other hand, washing increased insoluble fiber (+7%) and fat (+3%) contents. Concerning freezing, it did not change mango peels' nutritional composition, excluding protein and fat content, which were 27 % and 24 % lower in FZ than in W mango peels, respectively.

### 3.2. Phenolic compounds

Total and individual phenolic compounds identified in FPC, BBFC, and ABFC extracts from UNP, W, and FZ can be seen in Table 2. The yields of total phenolic compounds in methanolic extraction, basic hydrolysis, and acid hydrolysis were 192.45, 16.55, and 3.07 mg/100 g of FW in the UNP sample, 208.41, 18.10, and 3.73 mg/100 g of FW in W sample and, 184.69, 7.47, and 2.30 mg/100 g of FW in FZ samples, respectively. Hence, the results showed that the main quantity of phenolic compounds (free) was released from mango peels through methanolic extraction since their content in FPC was much higher than in BBPC (12–25 times) and ABPC extract (56–80 times).

Regarding phenolic profiles, thirteen different compounds belonging

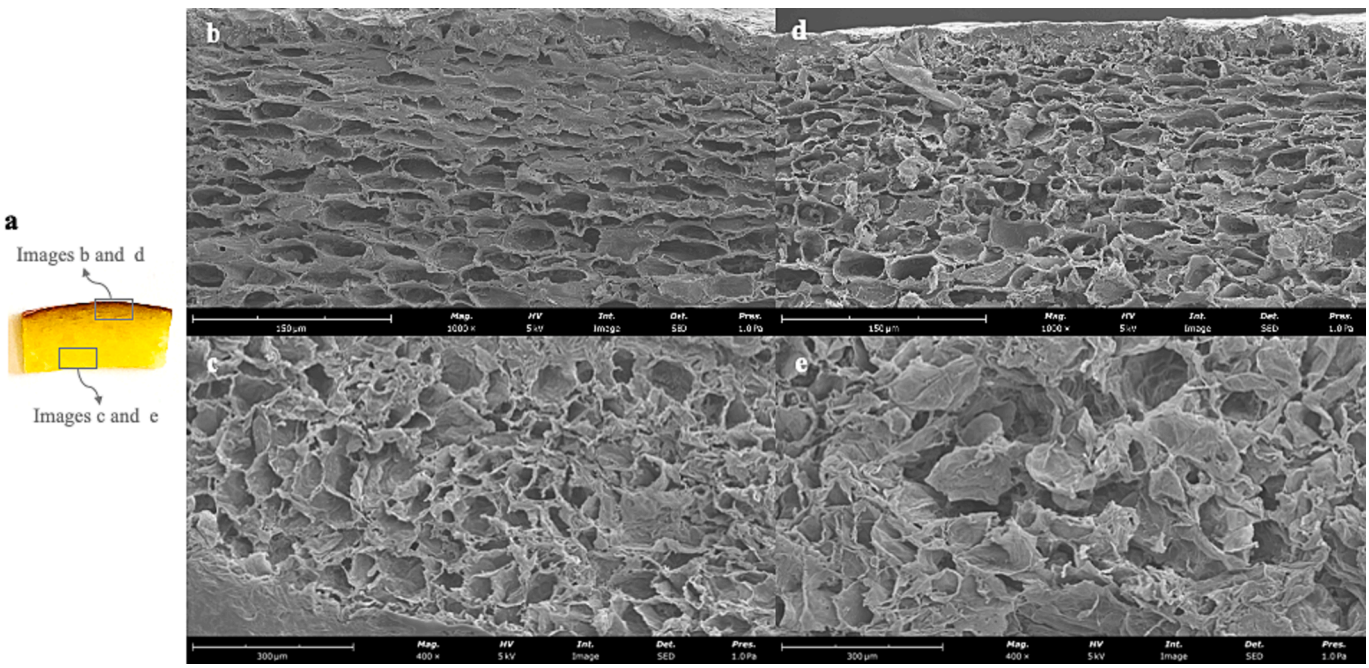


**Fig. 1.** Microstructure of washed (micrographs b and c) and frozen (micrographs d and e) mango peels observed through scanning electron microscopy (SEM). Image a depicts the cut of mango peels for SEM analysis and the regions of samples to which each image corresponds. The scale bars represent 150 (b and d) and 300 μm (c and e).

**Table 1**
Effect of washing and freezing on mango peels' nutritional composition.

| | | Dry Weight g/100 g FW | Total Carbohydrates g/100 g FW | Total fiber g/100 g FW | Soluble fiber g/100 g FW | Insoluble fiber g/100 g FW | Protein g/100 g FW | Fat g/100 g FW | Ash g/100 g FW | Vitamin C mg/g 100 FW |
|---|---|---|---|---|---|---|---|---|---|---|
| UNP | | 18.36 ± 1.60 | 16.73 ± 1.64 | 5.87 ± 0.24 | 3.06 ± 0.22 | 2.79 ± 0.03 | 0.90 ± 0.06 | 0.33 ± 0.00 | 0.37 ± 0.08 | 5.20 ± 0.17 |
| W | | 16.83 ± 0.68 | 15.13 ± 0.61 | 5.69 ± 0.17 | 2.76 ± 0.10 | 2.98 ± 0.06* | 0.99 ± 0.13 | 0.34 ± 0.00* | 0.35 ± 0.05 | 4.82 ± 0.03* |
| FZ | | 16.51 ± 1.04 | 15.11 ± 0.88 | 5.60 ± 0.17 | 2.90 ± 0.10 | 2.69 ± 0.39 | 0.72 ± 0.09# | 0.26 ± 0.01# | 0.42 ± 0.07 | 4.57 ± 0.16 |
| *p* value | UNP × W | 0.20 | 0.19 | 0.57 | 0.06 | 0.00 | 0.36 | 0.04 | 0.69 | 0.02 |
| | W × F | 0.68 | 0.97 | 0.75 | 0.21 | 0.18 | 0.04 | <0.00 | 0.22 | 0.11 |

Each value was expressed as mean ± standard deviation (n = 3). * indicates statistically significant differences between unprocessed and washed samples while # corresponds to statistically significant differences between washed and frozen samples ($p < 0.05$). FW – fresh weight; UNP – unprocessed sample; W – washed sample; FZ – frozen sample (at −20 °C for 30 days).

to five different families (xanthones; gallic acid and gallates; flavonoids; cinnamic acids; and other phenolic acids) were identified in the three samples. Cinnamic acids and other phenolic acids were only identified in BBPC and ABPC extracts, while gallates were only detected in FPC extracts. Flavonoids were identified in FPC and BBPC, whereas mangiferin and gallic acid were present in the three extracts.

The most abundant free phenolic compounds in all samples were mangiferin and gallic acid. They were followed by catechin or quercetin-3-O-galactoside, depending on the samples. However, the amount of quercetin-3-O-galactoside can be overestimated due to methodological limitations (Section 4.1). Regarding bound phenolic compounds, gallic acid was the most abundant compound in both extracts. Most of the bound phenolic compounds were released from mango peels during basic hydrolysis since a higher number of different phenolic compounds and higher concentrations of these bioactive compounds were identified in BBFC than in ABFC extracts.

Overall, washing did not affect bound phenolic compounds, excluding 4-hydroxybenzoic acid released from mango peels through acid hydrolysis. Its content was 54 % higher in W than in UNP samples. Regarding free phenolic compounds, washing caused a reduction in penta-O-galloyl-β-D-glucose (−23 %) and catechin (−30 %) contents, while the amounts of the other ones were preserved.

All free phenolic compounds tended to rise after freezing. However, only the increase of gallic acid (+36 %) and catechin (+51 %) had statistical significance. These results disagreed with values obtained for total phenolic compounds measured through the Folin-Ciocalteu assay since, although the difference was not statistically significant, total phenolic compounds tended to be lower in FZ samples than in W mango peels. On the other hand, freezing led to decreases in the amounts of total and all individual bound phenolic compounds, excluding mangiferin, quercetin-3-O-α-L-arabinofuranoside and ferulic acid. In the bound phenolic compounds released from mango peels through basic hydrolysis, the lowest and highest significant reductions corresponded to *p*-coumaric acid (−12 %) and gallic acid (−87 %). The other significant decreases were about 40 %, excluding 4-hydroxybenzoic acid (−57 %).

### 3.3. Carotenoids

Table 3 shows the carotenoids identified in UNP, W, and FZ mango peels and their quantification. β-carotene, lutein, and violaxanthin were identified in all samples. Regarding the presence of zeaxanthin, the results were not enlightening since it and lutein had the same retention time in the HPLC method used. This methodological limitation was detailly discussed in Section 4.1. The most abundant carotenoid in all samples was β-carotene, followed by lutein and violaxanthin.

The three identified carotenoids showed different susceptibilities to washing and freezing. Washing led to a decrease in lutein (−24 %), while freezing caused a reduction in violaxanthin content (−51 %). On the other hand, no differences were found between the β-carotene content of UNP and W and W and FZ samples.

### 3.4. Antioxidant activity

Table 4 presents the capacity to scavenge the ABTS and DPPH-free radicals of all studied phenolic extracts. In both assays, FPC extracts showed a much higher antioxidant activity than BBPC (ABTS assay: 12–31 times; DPPH assay: 11–32) and ABPC extracts (ABTS assay: 60–108 times; DPPH assay: 82–162).

Washing had no effects on the antioxidant activity of free (FPC extract) and bound phenolic compounds (BBPC and ABPC extracts). On the other hand, freezing caused a marked decrease in BBPC and ABPC extracts' capacity to scavenge the ABTS and DPPH-free radicals. These reductions were more accentuated in BBPC (ABTS: −72 %; DPPH: −70 %) than in ABPC extract (ABTS: −51 % and DPPH: −55 %). Concerning free phenolic compounds, differences between FPC extracts from W and FZ samples were not found.

### 3.5. Microstructure

Fig. 1 depicts how mango peels were cut to perform SEM analysis (image a), which sections of samples were observed (image a), and the microstructure of washed (micrographs b and c) and frozen mango peels (micrographs d and e). The comparison of images b and c or d and e showed that the microstructure of the epicarp and mesocarp closest to it was more compact and smoother than the mesocarp. In other words, as the images moved away from the epicarp, in both samples, the pores became larger and more irregular. It is important to consider that the scale bars of epicarp (b and d) and mesocarp micrographs (c and e) were non-identical (150 μm and 300 μm, respectively), so the difference in pore size was higher than the interpretation of the images without taking this discrepancy into account showed.

The impact of freezing on mango peels' microstructure was notorious, as it was observed that pores became larger or even ruptured (micrographs d and e). These changes were more exacerbated in the mesocarp (micrograph e) than in the epicarp (micrograph d).

## 4. Discussion

### 4.1. Nutritional composition, bioactive compounds, and antioxidant activity of mango peels

Overall, the macronutrients, ash, and vitamin C contents found in this study were within the ranges of values reported by previous studies (Kaur et al., 2023; Marçal & Pintado, 2021) (Section 3.1).

The free phenolic compounds of mango peels have been extensively studied. On the other hand, few research works extracted and identified bound phenolic compounds from mango peels and other vegetal

**Table 2**
Effect of washing and freezing on free and bound phenolic compounds released from mango peels through methanolic extraction, basic and acid hydrolyses, respectively.

| | Methanolic extraction µg/g FW | | | | | Basic hydrolysis µg/g FW | | | | | Acid hydrolysis µg/g FW | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | UNP | W | FZ | *p* value | | UNP | W | FZ | *p* value | | UNP | W | FZ | *p* value | |
| | | | | UNP × W | W × FZ | | | | UNP × W | W × FZ | | | | UNP × W | W × FZ |
| Total phenolic compounds | 1924.51 ± 243.15 | 2084.09 ± 361.42 | 1846.91 ± 146.99 | 0.56 | 0.35 | 165.45 ± 37.03 | 181.02 ± 45.55 | 74.68 ± 11.85# | 0.67 | 0.02 | 30.71 ± 5.13 | 37.30 ± 2.02 | 22.95 ± 1.87# | 0.11 | <0.00 |
| Mangiferin | 151.34 ± 11.45 | 162.20 ± 19.63 | 167.80 ± 11.44 | 0.45 | 0.69 | 7.76 ± 0.85 | 5.78 ± 0.98 | 4.97 ± 0.49 | 0.06 | 0.27 | 1.92 ± 0.12 | 1.55 ± 0.23 | 1.20 ± 0.18 | 0.07 | 0.11 |
| Gallic acid | 109.19 ± 7.31 | 102.78 ± 8.82 | 139.30 ± 10.07# | 0.39 | 0.01 | 52.50 ± 0.50 | 89.71 ± 17.03 | 11.30 ± 3.57# | 0.06 | 0.01 | 12.47 ± 4.58 | 15.62 ± 4.23 | 5.26 ± 0.78# | 0.43 | 0.01 |
| Penta-O-galloyl-β-D-glucose | 87.21 ± 2.71 | 67.52 ± 8.42* | 75.09 ± 2.67 | 0.02 | 0.21 | nd | nd | nd | – | – | nd | nd | nd | – | – |
| Methyl gallate | 53.70 ± 4.32 | 54.51 ± 9.11 | 55.23 ± 8.03 | 0.90 | 0.92 | nd | nd | nd | – | – | nd | nd | nd | – | – |
| Catechin | 97.98 ± 4.77 | 68.89 ± 3.34* | 103.92 ± 7.19# | <0.00 | 0.00 | nd | nd | nd | – | – | nd | nd | nd | – | – |
| Quercetin-3-O-galactoside[a] | 73.35 ± 5.26 | 82.63 ± 11.09 | 88.65 ± 4.13 | 0.26 | 0.47 | 3.72 ± 0.36 | 3.36 ± 0.30 | 2.02 ± 0.34# | 0.26 | 0.01 | nd | nd | nd | – | – |
| Quercetin 3-β-D-glucoside | 31.34 ± 1.77 | 30.81 ± 2.81 | 37.08 ± 3.64 | 0.80 | 0.08 | 3.50 ± 0.30 | 3.45 ± 0.74 | 2.07 ± 0.40# | 0.92 | 0.05 | nd | nd | nd | – | – |
| Quercetin 3-O-α-L-arabinopyranoside | 12.96 ± 0.80 | 13.33 ± 1.93 | 15.50 ± 1.31 | 0.77 | 0.18 | 1.27 ± 0.05 | 1.27 ± 0.26 | 0.80 ± 0.15# | 0.98 | 0.05 | nd | nd | nd | – | – |
| Quercetin-3-O-α-L-arabinofuranoside | 8.16 ± 0.52 | 8.76 ± 1.30 | 9.84 ± 0.88 | 0.50 | 0.30 | 0.86 ± 0.09 | 0.82 ± 0.17 | 0.55 ± 0.09 | 0.71 | 0.07 | nd | nd | nd | – | – |
| Ferulic acid | nd | nd | nd | – | – | 0.98 ± 0.01 | 1.06 ± 0.22 | 0.86 ± 0.05 | 0.58 | 0.20 | nd | nd | nd | – | – |
| p-Coumaric acid | nd | nd | nd | – | – | 2.29 ± 0.08 | 2.24 ± 0.01 | 1.97 ± 0.08# | 0.37 | 0.01 | 0.33 ± 0.04 | 0.34 ± 0.02 | 0.33 ± 0.02 | 0.92 | 0.89 |
| 3,4-Dihydroxybenzoic acid | nd | nd | nd | – | – | 7.83 ± 1.10 | 8.01 ± 0.58 | 5.07 ± 0.88# | 0.81 | 0.01 | nd | nd | nd | – | – |
| 4-Hydroxybenzoic acid | nd | nd | nd | – | – | 13.53 ± 0.92 | 11.15 ± 1.63 | 4.84 ± 1.01# | 0.09 | 0.01 | 2.83 ± 0.34 | 4.35 ± 0.25* | 1.27 ± 0.23# | 0.00 | <0.00 |

Total phenolic compounds were determined through Folin-Ciocalteu assay and were expressed in µg of gallic acid equivalents/g of fresh weight (FW). Each value was expressed as mean ± standard deviation (n = 3). * indicates statistically significant differences between unprocessed and washed samples while # corresponds to statistically significant differences between washed and frozen samples ($p < 0.05$). [a] In the HPLC method used quercetin-3-O-galactoside and ellagic acid had the same retention time, so these values can correspond to these both compounds. UNP – unprocessed sample; W – washed sample; FZ – frozen sample (at −20 °C for 30 days); nd – not detected.

**Table 3**
Effect of washing and freezing on mango peels' carotenoids.

| | | β-carotene μg/g FW | Lutein[a] μg/g FW | Violaxanthin μg/g FW |
|---|---|---|---|---|
| UNP | | 21.62 ± 1.65 | 1.19 ± 0.11 | 0.57 ± 0.04 |
| W | | 19.64 ± 0.08 | 0.90 ± 0.09* | 0.55 ± 0.01 |
| FZ | | 17.87 ± 1.59 | 0.75 ± 0.15 | 0.27 ± 0.02# |
| *p* value | UNP × W | 0.11 | 0.02 | 0.45 |
| | W × FZ | 0.13 | 0.22 | <0.00 |

Each value was expressed as mean ± standard deviation (n = 3). * indicates statistically significant differences between unprocessed and washed samples while # corresponds to statistically significant differences between washed and frozen samples ($p < 0.05$). [a] In the HPLC method lutein and zeaxanthin had the same retention time, so these values can correspond to these both compounds. FW – fresh weight; UNP – unprocessed sample; W – washed sample; FZ – frozen sample (at −20 °C for 30 days).

**Table 4**
Impact of washing and freezing on antioxidant activity of free and bound phenolic compounds released from mango peels through methanolic extraction and basic and acid hydrolyses, respectively.

| | | | ABTS μg of Trolox eq./g FW | DPPH μg of Trolox eq./g FW |
|---|---|---|---|---|
| Free phenolic compounds | UNP | | 5334.27 ± 198.59 | 4896.08 ± 215.09 |
| | W | | 5144.60 ± 326.59 | 4784.77 ± 547.25 |
| | FZ | | 4724.19 ± 297.42 | 4232.73 ± 289.94 |
| | *p* value | UNP × W | 0.44 | 0.76 |
| | | W × FZ | 0.18 | 0.20 |
| Bound phenolic compounds (basic hydrolysis) | UNP | | 455.92 ± 116.00 | 303.18 ± 7.89 |
| | W | | 549.62 ± 137.75 | 436.07 ± 127.56 |
| | FZ | | 153.67 ± 39.50# | 132.28 ± 32.81# |
| | *p* value | UNP × W | 0.21 | 0.15 |
| | | W × FZ | 0.00 | 0.02 |
| Bound phenolic compounds (acid hydrolysis) | UNP | | 76.92 ± 11.26 | 52.38 ± 8.13 |
| | W | | 85.84 ± 15.51 | 58.37 ± 8.73 |
| | FZ | | 43.70 ± 5.13# | 26.08 ± 5.62# |
| | *p* value | UNP × W | 0.47 | 0.43 |
| | | W × FZ | 0.01 | 0.01 |

Each value was expressed as mean ± standard deviation (n = 3). * indicates statistically significant differences between unprocessed and washed samples while # corresponds to statistically significant differences between washed and frozen samples ($p < 0.05$). eq. – equivalents; FW – fresh weight; UNP – unprocessed sample; W – washed sample; FZ – frozen sample (at −20 °C for 30 days).

byproducts (Dzah et al., 2020). According to Gómez-Caravaca et al. (2016) and López-Cobo et al. (2017), most phenolic compounds in mango peels were in free form, which agrees with what was observed in the present study (Section 3.2). As expected, considering the high discrepancies in the phenolic compound amounts, FCP expressed a much higher antioxidant activity than BBFC and ABFC extracts. The opposite happens in cereal matrices, namely corn, rice, and wheat, since bound phenolic compounds contribute to >70 % of total antioxidant activity (Adom & Liu, 2002). Furthermore, results showed that basic hydrolysis released most bound phenolic compounds from mango peels (Section 3.2). According to Pacheco-Ordaz et al. (2018), basic hydrolysis breaks esters bounds, releasing phenolic compounds linked to cell walls, while acid hydrolysis breaks glycoside bounds, releasing aglycones. Hence, the results suggested that most mango peels' bound phenolic compounds were attached to cell walls through esters bounds.

Regarding phenolic compounds profile, the most abundant families were xanthones; flavonoids; and gallic acid, gallates, and gallotannins, which agrees with the previously reported results (Ancos et al., 2018; Gómez-Caravaca et al., 2016; López-Cobo et al., 2017). Looking only for bound phenolic compounds, the present study and Pacheco-Ordaz et al. (2018) identified gallic acid as the most abundant phenolic compound, while in Gómez-Caravaca et al. (2016) and López-Cobo et al. (2017) studies ellagic acid was the most abundant one. The HPLC method used in the present study showed limitations regarding identifying ellagic acid since it and quercetin-3-O-galactoside had the same retention time. The peak to that retention time was identified as quercetin-3-O-galactoside because it had a UV absorption spectrum similar to the standard.

As in this study, Ruales et al. (2018) identified β-carotene, lutein, and violaxanthin in mango peels. The results of the present study were not enlightening regarding the presence of zeaxanthin. Lutein and zeaxanthin are stereo isomers, which difficult their identification and quantification separately (Boon et al., 2010). In the HPLC method used, lutein and zeaxanthin had the same retention time. Hence, although the peak identified as lutein had a UV absorption spectrum equal to its standard, it cannot be stated with certainty that zeaxanthin is not present and that the lutein amount is not overestimated. Ruales et al. (2018) and this research work used peels from the same mango cultivar and the same method to extract carotenoids. However, the contents reported by Ruales et al. (2018) were quite lower than those shown here. They used freeze-dried samples, which can contribute to reducing the values (Ancos et al., 2018). Furthermore, instead of β-carotene, they identified lutein as the most abundant carotenoid in mango peels (Ruales et al., 2018). On the other hand, β-carotene was the main carotenoid found in mango pulp, and its content was much higher there than in peel (Ruales et al., 2018).

The review of previous studies showed that the amounts of individual bioactive compounds detected in mango peels varied greatly (Marçal & Pintado, 2021). Discrepancies in cultivation conditions (e.g., soil composition, hydric stress, climate, sun exposure), ripening stage, and postharvest handling have been pointed out as the main reasons (Marçal & Pintado, 2021). Fig. 1a depicts the composition (epicarp and mesocarp proportion) of the mango byproducts used in the present study. Usually, the automatic peelers used by fruit processing companies do not enable the removal of only the epicarp (peel). Hence, the mango peels produced in huge amounts by mango processing companies, usually beside the epicarp, also include the mesocarp (pulp) closest to it. Different peeling processes and, consequently, different epicarp and mesocarp proportions could also contribute to this variability since peel and pulp have distinct chemical compositions (Gómez-Caravaca et al., 2016; López-Cobo et al., 2017; Ruales et al., 2018).

### *4.2. Impact of washing on nutritional composition, bioactive compounds, and antioxidant activity*

In recent decades, SH has been the most used disinfectant to wash fresh vegetal foods. However, SH is not recommended nowadays since it can react with organic matter, forming carcinogenic and mutagenic compounds (Hopkins et al., 2021). Marçal et al. (2022) showed that PAA is a sustainable alternative to SH regarding mango peels washing. Hence, in the present research work, mango peels were washed with PAA following the optimal conditions determined by Marçal et al. (2022). This washing procedure significantly improved mango peels' microbiological quality (Marçal et al., 2022).

Furthermore, in both present and Marçal et al. (2022) studies, these washing conditions enabled the preservation of mango peels' protein, ash, and total free phenolic compounds and their capacity to scavenge the ABTS and DPPH-free radicals (Sections 3.1, 3.2, and 3.4). However, in the present study, a more in-depth characterization of washed mango

peels was performed, and significant reductions in some antioxidant compounds, namely vitamin C, free phenolic compounds (penta-O-galloyl-β-D-glucose and catechin), and carotenoids (lutein) were detected (Sections 3.1, 3.2, and 3.3). On the other hand, an increase in insoluble nutrients such as insoluble fiber and fat was observed (Section 3.1). The reductions mentioned above occurred due to the leaching and oxidation reactions during washing since PAA is an oxidizing agent. Consequently, these losses led to a concentration of insoluble nutrients.

To our best knowledge, until now, excluding Marçal et al. (2022), there are no available studies about the impact of washing with PAA on the nutritional composition and bioactive compounds of vegetal byproducts. Even the effect of disinfection with PAA on edible fruits and vegetables' chemical composition has been little studied. Vandekinderen et al. (2009) studied the vitamin C retention on ready-to-eat white cabbage washed with 0, 80, and 250 mg/mL of PAA (food product to disinfectant solution ratio: 1:10 (kg:L); disinfection time: 5 min) and they observed a reduction of 24.87 %, 37.93 %, and 45.57 %, comparing with unwashed samples. Moreover, they reported that washing with 80 and 250 mg/mL of PAA decreased the β-carotene content by 49 % and 46 %, respectively, while no statistically significant reductions in lutein amount were detected. Velde et al. (2016) showed that fogging with PAA at concentrations between 3.4 and 116.6 μl/L for 5.7–69.3 min can marked change the strawberries' phenolic compounds profile. The phenolic losses varied depending on PAA concentration, disinfection time, and the chemical structure of phenolic compounds (de Velde et al., 2016).

Disinfectant washing applied slightly impaired mango peels' phytochemical composition. In fruit processing industries, mangoes normally are disinfected before being peeled. However, if the peels are not processed quickly, foodborne and spoilage microorganisms can overgrow, compromising mango peels' food safety and nutritional value. Furthermore, it is important to consider that some microorganisms survive in processing steps like freezing and drying, namely the sporulating ones. Hence, washing is usually an essential step in mango peels processing to obtain safe functional ingredients.

### 4.3. Impact of freezing on nutritional composition, bioactive compounds, antioxidant activity, and microstructure

#### 4.3.1. Nutritional composition, bioactive compounds, antioxidant activity

As far as authors know, this study described the effect of freezing on mango peels' phytochemical composition and antioxidant activity for the first time. Regarding the impact on nutrients, the decrease in protein and fat content and vitamin C preservation stood out (Section 3.1). Vitamin C preservation is an important advantage since mango peels have a significant amount of this micronutrient, and its intake is essential for bodily functions. Several research works reported reductions in fruit products' vitamin C content after frozen storage (Bonat Celli et al., 2016). Water loss during freezing, frozen storage, and thawing (due to freezing concentration and drip loss) was pointed out as the reason for vitamin C reduction since it is quite hydrophilic and, therefore, easily carried by water (Bonat Celli et al., 2016). In this study, mango peels were still frozen when they were milled to subsequently extract vitamin C and other bioactive compounds, avoiding the negative impact of drip losses during thawing (Section 2.3).

Concerning the effect of freezing on the carotenoids profile, a marked reduction in violaxanthin content was detected (Section 3.3). In both processing steps, washing and freezing, it is unlikely that carotenoids losses have occurred due to leaching and drip losses since they are extremely hydrophobic (Boon et al., 2010). Oxidation reactions, epoxidation, and isomerization are the more plausible reasons for carotenoids reduction (Bonat Celli et al., 2016).

The results regarding the impact of freezing on free phenolic compounds obtained through Folin–Ciocalteu and HPLC methods were discrepant. The content of all individual free phenolic compounds increased after freezing, while total free phenolic compounds tended to be lower in FZ than in W samples (Section 3.2). Folin–Ciocalteu is an electron transfer-based assay widely used to determine total phenolic compounds in foods. However, it has some limitations since, besides phenolics, other compounds with reducing powder (e.g., some amino acids and proteins, fructose, sucrose, and organic acids) can overestimate the results (Margraf et al., 2015). So, the lower value of total free phenolic compounds detected in FZ samples than in washed ones probably resulted from the decrease of other compounds with reducing capacity, namely proteins.

The increase and the marked reduction of free and bound phenolic compounds resulted from the changes in mango peels' microstructure (Sections 3.2, 3.5, and 4.3.2). The destruction of cell walls by ice crystal growth improved the extractability of these bioactive compounds and converted some bound phenolic into free ones. Thus, freezing can be a very advantageous preservation method when the main purpose of mango peels processing is to produce phenolic extracts. The positive effect of freezing on phenolic compounds' extractability was previously observed in other raw materials (Bonat Celli et al., 2016; Dzah et al., 2020). For instance, Oliveira et al. (2016) reported an increase in the catechin content (+47 %) from peaches stored at −20 °C for 360 days. However, the same study found significant decreases in other free phenolic compounds, namely chlorogenic acid (−39 %). Dzah et al. (2020) proposed that in the future, slow-freezing-thawing processes will be explored as a means to increase the extractability of bound phenolic compounds from vegetal byproducts.

The increase of free phenolic compounds after freezing did not result in a rise in FPC extracts' antioxidant activity, possibly due to losses of other antioxidant compounds (e.g., violaxanthin). However, marked reductions in both bound phenolic extracts' antioxidant activity were observed, suggesting that freezing could change the health benefits of mango peels' fiber (Section 3.4). During digestion, bound phenolic compounds remain linked to fiber until they reach the colon, conferring it antioxidant properties (Angulo-López et al., 2022). Furthermore, some phenolic compounds can exert prebiotic activity (Zhang et al., 2023). Both prebiotic and antioxidant activities promote colon health. Studying the impact of freezing on gastrointestinal digestion, bioaccessibility, bioavailability, and prebiotic activity of phenolic compounds from mango peels represents one opportunity for future research. Freezing could increase phenolic compounds' bioaccessibility and bioavailability, promoting their systemic health benefits. On the other hand, this processing method can damage phenolic compounds' resistance to gastric conditions, antioxidant activity in the colon, and prebiotic properties.

The present study showed that freezing (according to the conditions applied here) is an adequate method to preserve mango peels. However, the incorporation of mango peel pulps into foods was little explored until now, being necessary to study its impact on products' sensory properties, preservation, nutritional value, and health benefits (Marçal & Pintado, 2021). Using freezing as a complementary method or pretreatment to drying, namely hot air drying, also represents one opportunity for future research. The damage in cell walls caused by ice crystals growth could facilitate water evaporation, decreasing the drying time. On the other hand, it could reduce the resistance of some bioactive compounds to the applied conditions since they are less "protected" by fibers and other macromolecules.

#### 4.3.2. Microstructure

To our best knowledge, this was the first study to evaluate the effect of freezing on mango peels' microstructure, using SEM (Section 3.5). However, the impact of freezing and storage at different temperatures (−80 and 18 °C for six months) on peeled mangoes' microstructure was previously evaluated by Li et al. (2020). The changes that they observed in mangoes frozen at −18 °C compared with fresh ones were similar to those shown in micrographs **e** and **c** (Fig. 1), respectively. In both studies, it was observed that frozen samples had larger or ruptured pores, which suggests tissue damage caused by ice crystals growth (Li

et al., 2020). The present study showed that the cellular tissue destruction caused by freezing was higher in the mesocarp (image **e**) than in the epicarp (image **d**), probably because their cell wall structure was weaker. Lopes et al. (2016) performed a histological analysis of mangoes' epicarp and mesocarp using a light microscope. They observed that cell walls in mesocarp were less rigid and thinner than in epicarp (Lopes et al., 2016).

Li et al. (2020) also reported that increased storage time led to higher tissue damage. During storage, the number of ice crystals decreases while the size of the remaining ones increases. This phenomenon occurs due to recrystallization mechanisms (Kumar et al., 2019). In the present work, mango peels were submitted only to 24 h of frozen storage to fix, dehydrate, and dry (Section 2.9) the washed and frozen samples simultaneously. It is important to highlight that freezing was the only variable leading to microstructure changes. However, if the microstructure of mango peels had been analyzed after 30 days of frozen storage (conditions applied to evaluate the other parameters), probably greater tissue destruction would have been observed.

The pieces used to compare the W and FZ samples' microstructures were from the same mangoes. The effect of freezing on foods' microstructure is conditioned not only by freezing conditions but also by sample characteristics (Li et al., 2020; Siramard & Charoenrein, 2014). For instance, Siramard and Charoenrein (2014) reported greater tissue damage in ripe mangoes than in partially ripe ones.

## 5. Conclusion

This study showed that mango peels are an excellent source of nutrients (fiber and vitamin C) and bioactive compounds, namely mangiferin, gallic acid, and β-carotene, related to the prevention of several diseases. To our best knowledge, this research work described the impact of freezing on mango peels' phytochemical composition and microstructure for the first time. Additionally, the effect of washing with PAA on the mango peels' fiber, vitamin C, phenolic compounds profile, and carotenoids profile has not been evaluated until now.

Washing slightly impaired mango peels' phytochemical composition since it decreased the content of vitamin C (−7%), penta-O-galloyl-β-D-glucose (−23 %), catechin (−30 %), and lutein (−24 %). However, taking into account that antioxidant activity, fiber, and main bioactive compounds were preserved, washing with PAA can be considered an adequate method to decrease mango peels' organic and microbial loads.

Freezing caused marked changes in mango peels' microstructure. The damage of cell walls led to an increase in free phenolic compounds, namely acid gallic (+36 %) and catechin (+51 %), and a drastic reduction in bound phenolic compounds. Regarding antioxidant activity, it was preserved in free phenolic compounds extracts but markedly reduced in bound phenolic compounds extracts. Additionally, freezing decreased the amount of violaxanthin (−51 %). However, the main nutrient (fiber) and the most abundant bioactive compounds remained unchanged or increased in frozen mango peels, suggesting that overall freezing is an adequate method to preserve mango peels. Its application can be particularly advantageous in the production of phenolic extracts. Nevertheless, other preservation methods should be considered when the desired final product is fiber with antioxidant properties.

## Funding

This work was supported by the National Funds from FCT - Fundação para a Ciência e Tecnologia through project UIDB/50016/2020 and by FCT individual PhD grant (SFRH/BD/145301/2019).

## CRediT authorship contribution statement

**Sara Marçal:** Writing – original draft, Visualization, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Sérgio Sousa:** Methodology, Investigation, Formal analysis. **Helena Araújo-Rodrigues:** Methodology, Investigation. **Inês V. Silva:** Methodology, Investigation, Formal analysis. **Débora A. Campos:** Writing – review & editing, Validation, Supervision, Methodology. **Manuela Pintado:** Writing – review & editing, Validation, Supervision, Resources, Project administration, Methodology, Funding acquisition, Conceptualization.

## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Data availability

No data was used for the research described in the article.

## Acknowledgment

The authors thank Nuvi Fruits, S.A, for providing the mango peels.

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