



Iolanda Nicolau-Lapeña ^{1,*}, Jordi Ortiz ^{1,*}, Inmaculada Viñas ¹, Maribel Abadias ², Gloria Bobo ² and Ingrid Aguiló-Aguayo ²

- ¹ Food Technology Department, AGROTECNIO-CERCA Center, Universitat de Lleida, Rovira Roure 191, 25198 Lleida, Spain; inmaculada.vinas@udl.cat
- ² IRTA, Postharvest Programme, Parc Científic i Tecnològic Agroalimentari de Lleida, Parc de Gardeny, Edifici Fruitcentre, 25003 Lleida, Spain; isabel.abadias@irta.cat (M.A.); ingrid.aguilo@irta.cat (I.A.-A.)
- * Correspondence: iolanda.nicolau@irta.cat (I.N.-L.); jordi.ortiz@irta.cat (J.O.)

Abstract: The postharvest life of strawberries is short, and disinfection processes for fresh-cut and frozen strawberries are needed to address the risk posed by foodborne pathogens in this kind of product. For this, a process involving immersion in a 40 mg L^{-1} peracetic acid (PA) solution accompanied by the use of an emerging technology, ultraviolet-C for 2 min, was studied for its impact on strawberry quality and microbial load as a novel alternative method to chlorine sanitation. The shelf life of the washed strawberries was evaluated in fresh (whole or fresh-cut) product for 11 days at 4 $^{\circ}$ C and in product that had been frozen for 12 months at –20 $^{\circ}$ C (air or modified atmosphere, 20% CO₂, 5% O₂, and 75% N₂). After washing, total aerobic mesophylls, yeast, and mold decreased by 0.5–1.0 log units and these counts remained low during storage. The fresh and fresh-cut fruits' firmness (2.3 \pm 0.5 N at day 11) and lightness (expressed by L*, averaging 31.0 \pm 0.1) were maintained. Although antioxidant activity, expressed by the amount of radical 2,2-diphenyl-1-picrylhydrazyl (DPPH), decreased during storage in fresh-cut samples, total ferric reducing antioxidant power (FRAP), total phenolic content, and total anthocyanin content were maintained in all the formats during storage (averaging 1.33 ± 0.04 g kg⁻¹ ascorbic acid equivalents, 0.212 ± 0.01 g kg⁻¹ gallic acid equivalents, and 0.03 ± 0.01 g kg⁻¹ pelargonidine-3-glucoside). An increase in red color (from 32.1 to 39.3 a* values) and a loss of firmness of up to 46.8% was observed after the first month of frozen storage with no changes in the nutritional quality. Considering the sanitizing effect of water UV-C with peracetic acid (WUVPA) and the results obtained in the present study, the addition of this process in the production chain of strawberries could be an effective method to maintain the shelf life of the fruits, especially for fresh-cut strawberries.

Keywords: fruit; disinfection; storage; microbiological load; vitamin C

1. Introduction

Strawberries (*Fragaria* × *ananassa*) are widely consumed because of their excellent organoleptic properties, high bioactive compound content, and high antioxidant content [1]. However, their postharvest life is short due to their relatively high metabolic activity, sensitivity to fungal decay, and susceptibility to water loss. Also, they are susceptible to mechanical injuries owing to their soft texture and lack of a protective rind [2]. As such, the storage and selling of these fruits is very challenging in domestic and international markets, and spoilage or deterioration sometimes occurs before the product even reaches the consumers [3]. If not solved, actual product loss—10% to 35% at retail and at consumer levels, respectively—can have a great impact, causing economic losses in the sector [4].



Citation: Nicolau-Lapeña, I.; Ortiz, J.; Viñas, I.; Abadias, M.; Bobo, G.; Aguiló-Aguayo, I. Ultraviolet-C Light and Peracetic Acid Extend the Shelf Life of Fresh and Frozen Strawberries. *Horticulturae* 2024, *10*, 452. https:// doi.org/10.3390/horticulturae10050452

Received: 27 February 2024 Revised: 21 April 2024 Accepted: 23 April 2024 Published: 29 April 2024



Copyright: © 2024 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/).

The necessity of devising novel approaches for controlling postharvest diseases has arisen alongside increasing consumer awareness regarding pesticide residues on food, pathogen resistance, and nutritional losses [5]. Postharvest handling procedures can pose challenges with regard to strawberries, particularly due to their susceptibility to mechanical damage. Refrigeration at 4–5 $^{\circ}$ C or freezing (–20 $^{\circ}$ C), either alone or in conjunction with a modified atmosphere, has demonstrated efficacy in slowing down metabolic processes and extending the shelf life of strawberries [6]. Fresh strawberries are often packaged directly in the field and sold without undergoing any sanitation process. Conversely, freshcut and frozen strawberries typically undergo washing and/or disinfection steps before commercialization. Regardless of the method, these fruits are consumed raw, underscoring the imperative of providing consumers with safe products, thereby driving the need for the development of new sanitization strategies. Chlorine is widely utilized as a disinfectant in the fruit industry owing to its affordability, ease of use, and effectiveness against certain vegetative bacteria and enteric viruses [7]. However, its efficacy is highly dependent on pH levels, and it reacts with organic matter, resulting in the formation of undesirable by-products such as trihalomethanes. Consequently, efforts are underway to explore alternative products or technologies that offer effective substitutes for chlorine. Frozen strawberries have also been linked to safety issues associated with foodborne pathogens, such as Salmonella spp. and human norovirus [8]. The importance of maintaining their safety lies in the fact that these products are not only consumed at home but are also used as an ingredient in a variety of groceries, including yoghourts, smoothies, and ice creams [9].

Different alternatives have been studied to reduce microbial growth and to extend the shelf life of strawberries. In this regard, the combination of water-assisted ultraviolet C light (WUV-C) and peracetic acid (PA) has proven effective in reducing *Listeria monocytogenes* and *Salmonella enterica* populations in whole strawberries, and it did not negatively affect the quality and nutritional parameters of the fruit [10]. Moreover, in previous studies by our group, the combination of WUV-C and PA at 40 mg L⁻¹ reduced artificially inoculated norovirus in strawberries reaching to 2 log Tissue Culture Inhibition Dose at 50% or TCID₅₀ g⁻¹. *L. monocytogenes* and *S. enterica* artificially inoculated in strawberry were also reduced reaching to 4.5 and 2.8 log CFU g⁻¹ after the washing treatments [11].

Therefore, in this study, the combination of WUV-C and 40 mg L^{-1} of PA for 2 min was suggested as a disinfectant step in the processing chain of strawberries and was used to increase the shelf life of this product in three different formats: fresh, fresh-cut, and frozen strawberries. For this, quality parameters (including pH, TSS, and TA, color, and firmness evolution), antioxidant capacity, phytochemical changes (total phenolic, total anthocyanin, or total ascorbic acid retention), and the evolution of the epiphytic microbiota in strawberries were evaluated.

2. Materials and Methods

2.1. Materials

Strawberries (*Fragaria* \times *ananassa*) cv. San Andreas were purchased from local providers (Spain) during the campaign in 2019 and stored at 4 °C until use. The strawberries were used the same day they were bought.

Yeast extract, plate count agar (PCA), dichloran rose bengale chloramphenicol agar (DRBC), and peptone were obtained from Biokar Diagnostics (Allonne, France). Peracetic acid (PA) 15% was purchased from Panreac AppliChem (Barcelona, Spain).

Ascorbic and gallic acids, including 2,4,6-tris(2-pyridyl)-s-triazine (TPTZ), 2,2-diphenyl-1-picrylhydrazyl (DPPH), sodium carbonate, metaphosphoric acid, acetic acid, and 3,3',3''phosphanetriyltripropanoic acid (TCEP), were acquired from Sigma-Aldrich (Steinheim, Germany). Methanol, acetone, chlorohydric acid (37%), sodium acetate, sodium hydroxide, sodium chloride, potassium chloride, ferric chloride hexahydrate, and Folin Ciocalteau's reagent were procured by Panreac AppliChem (Barcelona, Spain).

2.2. Preparation of Disinfection Equipment

The effect of water-assisted UV-C (WUV-C) combined with 40 mg L⁻¹ peracetic acid (PA) on the shelf life of strawberries was studied in two formats: fresh and frozen. Treatments were conducted in the UV-C water-assisted (WUV-C) equipment LAB-UVC-Gama (UVC-Consulting Peschl España, Castellón, Spain), as already described by Nicolau-Lapeña et al. [10] and Ortiz-Solà et al. [11]. Before the experiment, the 4 UV-C lamps were preheated for 10 min. After this time, the irradiance value in the empty tank, which was measured with an Easy H1 UV sensor (Peschl Ultraviolet, Mainz, Germany) through an orifice located on the lid of the tank, was $10.5 \pm 0.5 \text{ W/m}^2$. Afterwards, the WUV-C tank was filled with 12 L of cold (6 ± 2 °C) water and the UV-C lights were switched on for 5 min. When needed, PA or sodium hypochlorite were added in the tank.

Water parameters, including pH, oxidation-reduction potential (ORP), and turbidity, were measured before and after each treatment. ORP and pH were measured using a GLP22 Crison, Alella pH meter (Barcelona, Spain) equipped with a pH probe (ref. 52-03, Crison) or ORP probe 62-51 Hach (Geneva, Switzerland), respectively. ORP was measured in millivolts (mV). Turbidity was measured using a portable turbidimeter (TN-100, Eutech, Singapore) measuring in nephelometric turbidity units (NTU).

2.3. Disinfection Treatments

Before the treatment, peduncles and leaves were carefully removed. For fresh presentation, whole strawberries were washed using WUV-C 4 lamps with 40 mg L⁻¹ PA for 2 min (WUVPA, W-TREAT). Tap water applied for 2 min was used as a control (W-CON). A lot of the strawberries washed under WUV-C 4 lamps with 40 mg L⁻¹ PA for 2 min were freshly cut (C-TREAT) in half with an average weight of 12.5 ± 0.2 g. For fresh-cut strawberries, sodium hypochlorite (NaOCl) 200 mg L⁻¹ adjusted to pH 6.5 using citric acid 2 M at 6 ± 2 °C for 2 min was used as a control (C-CON). After NaOCl disinfection, strawberries were rinsed in tap water during 2 min. After washing, the fruits were left at room temperature to drain the excess water (1 h). Afterwards, 400 ± 10 g of strawberries were weighed in 1000 mL polypropylene (PP) trays (CL1000TPE, Alphacel) and sealed with a propylene HS 1/17 film (ACSA, Valencia, Spain). Packaged product was stored in a display case (INFRICO Z017ERC110, Córdoba, Spain) at 4.5 ± 0.4 °C for 11 days. Light and dark cycles were programed to 18 and 8 h, respectively, to simulate the opening and stocking hours of a supermarket display.

For the frozen presentation, whole strawberries were washed using WUV-C 4 lamps with 40 mg L⁻¹ PA for 2 min (WUVPA). After washing, the fruits were left at room temperature to drain the excess water (1 h). Strawberries (400 ± 10 g) were weighed in 1400 mL (MW1-50, Wiezoplast, Oss, The Netherlands) trays and sealed with a HS 1/17 film. Half the samples were packed in modified atmosphere packaging (MAP) (20% CO₂, 5% O₂, and 75% N₂), and the other half were packed in air. Packages with strawberries were frozen at -80 °C in a freezing cabinet with liquid nitrogen (N₂) (model Mini Batch CM-85/1090, Metal Carbide, Carburos Metalicos, Barcelona, Spain). The transition time was 40 min, and the internal temperature of strawberries reached -15 °C after 60 min (Figure 1). The strawberries were stored at -25.0 ± 0.5 °C for 12 months.



Figure 1. Freezing parameters and curves of strawberry samples during supercooling in a freezing cabinet functioning with liquid nitrogen (N_2), using the Mini Batch CM-85/1090 model from the brand Metal Carbide.

2.4. Quality of Strawberries

Fresh strawberries, whole or fresh-cut, were sampled at days 0, 2, 4, 7, and 11 (D0, D2, D4, D7, and D11). Quality parameters were determined each sampling day. Then, a proportion of the samples of each treatment were frozen with liquid nitrogen, milled using a MINIMOKA GR-020 grinder (Taurus Group, Barcelona, Spain), and stored at -80 °C for further biochemical analysis.

Frozen strawberries were sampled at months 0, 3, 6, and 12 (M0, M3, M6, and M12) and defrosted at 4 °C overnight. Quality determinations were performed on thawed product, and aliquots were prepared and stored at -80 °C for further biochemical analyses.

For each treatment and strawberry format (fresh, fresh-cut, and frozen), the strawberries (fully ripe) were submerged in the water tank for agitation at a ratio of 3:10 (strawberries:water, w:v). The number of sample replicates for each analytical study is detailed below. The experiment was performed twice to achieve homogeneity of the results.

2.4.1. Physiochemical Evaluation of Quality Parameters

To determine pH, total soluble solids, and titratable acidity, strawberries were smashed in a blender to prepare 25 mL of juice. Each parameter was evaluated twice for each repetition (n = 3) according to Nicolau-Lapeña et al. [12].

The respiration rate (RR) of fresh and fresh-cut strawberries was determined immediately after processing. For this, 100 ± 5 g strawberries were put inside a hermetic plastic pot and stored at 4 °C (n = 4). After 24 h, O₂ and CO₂ concentrations were measured using a CheckMate 3 headspace gas analyzer (Dansensor, Barcelona, Spain). RR was calculated using Equation (1):

$$RR (mL kg^{-1} h^{-1}) = \frac{[CO_2]_f - [CO_2]_i \cdot (V_t - V_0) \cdot 0.01}{W \cdot (t_f - t_i)}$$
(1)

where $[CO_2]_f$ – $[CO_2]_i$ is the change in concentration between measurements (% in volume), V_t is the total volume of the container (600 mL), V_0 is the volume of the fruits (mL) (considering their density and according to their weight), t_f – t_i is the time difference between measurements (h), and W is the weight of the fruits in the container (kg).

The evolution of internal gas composition in the sample trays was assessed by measuring O_2 and CO_2 concentration with CheckMate 3 (Dansensor, Spain) on each storage day (D) and was expressed in % (n = 3).

The color of 10 strawberries was measured on 3 sides using a CR-200 Minolta Chrome Meter (Minolta, Inc., Tokyo, Japan) with a D65 illuminant and a 10° observer angle. The instrument was calibrated using a standard white reflector plate. Color was expressed as CIE L* a* b* coordinates (n = 30).

Texture changes were evaluated in 10 samples for treatment and for testing. Two textural tests were carried out using the TA.XT Plus Connect texture analyzer (Stable Micro systems Ltd., Surrey, UK). Puncture tests were performed with a 4 mm cylindrical probe, measuring the maximum force encountered when the probe enters 8.0 mm deep into the tissue. Both tests were run at 5 mm s⁻¹ speed with a trigger force of 0.1 N.

Drip losses were determined by weighing the exudates immediately after the freezingthawing process. It was calculated by the difference in weight, as expressed in Equation (2):

$$DL(\%) = \frac{We}{Wf} \times 100$$
⁽²⁾

where *We* is the weight of the exudate and *Wf* is the weight of the frozen product.

2.4.2. Antioxidant Capacity

The antioxidant activity of strawberries was assessed by ferric reducing antioxidant power (FRAP) and DPPH scavenging activity assays, as described by Nicolau-Lapeña et al. [12]. Methanol 70% (*v:v*) with a retention time of 10 min (4 °C) was used for the extraction method. Results from 3 repetitions (n = 3) are expressed as ascorbic acid equivalents to a fresh weight basis, in g kg⁻¹.

2.4.3. Phytochemical Assays

The total phenolic content (TPC) was assessed by the Folin Ciocalteau method using the same extract used for antioxidant activity determination, following the procedure described by Nicolau-Lapeña et al. [12]. Results from 3 repetitions (n = 3) were expressed as gallic acid equivalents to a fresh weight basis, in g kg⁻¹.

For the total anthocyanin content (TAC), extracts and quantification were carried out in triplicate (n = 3) according to the method described by Meyers et al. [13]. Methanol 80% (v:v) was used for the extraction method. Anthocyanin content was expressed as cianidine-3-glucosyde according to a fresh weight basis, in g kg⁻¹.

Total ascorbic acid contents, expressed as the sum of ascorbic acid and dehydroascorbic acid (TAA), were determined by high-performance liquid chromatography (HPLC) in a Waters 717 plus Autosampler HPLC system (Waters Corp., Milford, USA) coupled to a UV detector, following the method described by [14]. Metaphosphoric acid 3% (*w*:*v*) and acetic acid 8% (*v*:*v*) with a retention time of 5 min (4 °C and darkness) were used for the extraction method. One extraction was performed per sample per repetition (*n* = 3). Average peak areas of duplicate injections were used for quantification. The concentration of vitamin C, expressed according to a fresh weight basis in g kg⁻¹, was calculated by area interpolation on the adequate calibration curve.

2.4.4. Microbiological Quality

The effect of the washing treatments on total aerobic mesophylls (TAM) and yeasts and molds (Y&M) was evaluated. For this, 25 g per repetition (n = 3), taken from pieces of 2 strawberries to ensure representativity, was diluted 1:4 in peptone buffered solution. The count process followed the method described in Nicolau-Lapeña et al. [12]. Results were

expressed as log colony-forming unit (CFU) per gram (CFU g^{-1}), and the detection limit was 1.30 log CFU g^{-1} .

2.5. Statistical Analysis

For fresh product, data were checked for significant differences by applying an analysis of variance test (ANOVA). The criterion for statistical significance was p < 0.05. When significant differences were observed, a contrast analysis was applied, comparing formats with the same treatment (W-TREAT vs. C-TREAT) and treatments within the same format (W-CON vs. W-TREAT and C-CON vs. C-TREAT). For frozen product, Student's *t*-test was run between the two gas composition conditions, and a Tukey HSD (honest significant difference) test was applied to evaluate differences between times within the same treatment. The criterion for statistical significance was p < 0.05. All statistical analyses were carried on using JMP 13 (SAS Institute Inc., Cary, NC, USA).

3. Results and Discussion

3.1. Working Solutions during Treatments

For water containing PAA at 40 mg L⁻¹, values were 5.1 ± 0.2 and 0.447 ± 0.013 V for pH and ORP, respectively. For water with chlorine at 200 mg L⁻¹, values were 6.7 ± 0.2 and 0.907 ± 0.011 V, respectively. The turbidity of the water (2.0 ± 0.1 NTU) was in agreement with the values observed in previous studies by this group, indicating that the absorbance of water at 254 nm (corresponding to that of the UV light) was low and no irradiation interference was caused by the presence of particles or dirt in the water [10].

3.2. Fresh and Fresh-Cut Strawberries

3.2.1. Commercial Quality of the Fruits

The washing treatments using WUVPA and the respective controls for whole and fresh-cut presentation formats were evaluated for different quality aspects during storage.

No differences were found in respiration rate (RR) between whole and fresh-cut samples or between the treatments, which is not in agreement with the common knowledge in the literature [15]. However, no reason for this behavior could be elucidated. RR was moderate, averaging 7.6 ± 0.4 mL CO₂ kg⁻¹ h⁻¹. Gas composition in the package showed the same evolution for all four treatments, achieving final values of $11.1 \pm 0.7\%$ O₂ and $12.3 \pm 0.9\%$ CO₂ (Table S4).

In terms of overall quality (pH, TSS, and TA values), after disinfection (D0), there were no significant differences between different treatments or presentations (Table 1). The results found were in accordance with those in the literature [16]. The pH values of samples after 11 days of storage ranged between 3.3 and 3.4. However, some variations during the storage period were observed. TSS decreased in W-CON- and C-CON-treated fruits (6.3 \pm 0.3 to 5.3 \pm 0.2%) after 11 days of storage (Table 1). This trend was also observed by Aday et al. [17] in strawberries during storage after sonication treatment (30–90 W, 5–10 min). Some authors attribute this trend to a reduction in fruit metabolism, resulting in lower decomposition of these constituents and a slow hydrolysis of sugars [18]. Nonetheless, in the present study, no differences were observed between the RR of the different treatments or formats. This fact could explain why the TSS changes only happened in one treatment. The results regarding the TA values of strawberries (Table 1) show that there was a decrease in this parameter during storage. According to Aday and Caner et al. [18], the use of organic acids in strawberry metabolic processes might be related to this decrease, which was faster in samples treated with WUVPA than it was in those that received the control treatment.

Regarding the color of the samples, L* (luminosity) and a* (redness) coordinates from the CIELab space did not differ between treatments and averaged 38.7 ± 2.2 and 32.5 ± 0.5 , respectively (Figure 2). These values indicate the characteristic red light color of strawberries as reported in the literature [19,20]. At the end of the storage period, luminosity was maintained and redness decreased regardless the treatment. The decrease in a* values was sharper for whole fruits than for cut samples. The decrease in a* could be attributed to the higher stress induced in the fresh-cut strawberries, which could accelerate browning reactions and the progressive loss of red color in the plant tissue. This trend was also reported by Aday and Caner et al. [18], who also found a decrease in redness in half-cut strawberries treated with ultrasound technology.

Table 1. Quality assessment of fresh and fresh-cut strawberries: pH, total soluble solids (TSS, %), titratable acidity (TA mg L⁻¹), firmness (N), and biochemical parameters: antioxidant activity, expressed by DPPH· and FRAP values (expressed as AAE according to FW basis, in g kg⁻¹); total phenolic content (TPC, expressed as GAE according to FW basis, in g kg⁻¹); total anthocyanin content (TAC, expressed as pelargonidin-3-glucosyde according to FW basis, in g kg⁻¹); and total ascorbic acid (TAA, ascorbic acid according to FW basis, in g kg⁻¹) in fresh strawberries. Values are the mean of three repetitions ± standard deviation (*n* = 10 in the case of firmness). Different lowercase letters show statistically significant differences (*p* < 0.05) between treatments within the same presentation (whole or fresh-cut): 'a' and 'b' are used for the fresh-cut fruit (analyzed with contrast test). Asterisks show statistically significant differences (*p* < 0.05) between presentation formats (whole vs. fresh-cut) for samples treated with water-assisted ultraviolet-C with peracetic 40 ppm. Capital letters show statistically significant differences (*p* < 0.05) over time within the same treatment analyzed with Tukey's test.

	Format	Treatment ¹	D0	D4	D7	D11
рН	Whole	W-CON W-TREAT	$3.5 \pm 0.1 \ ^{aB}$ $3.4 \pm 0.1 \ ^{aAB}$	$3.5 \pm 0.1 \ ^{aB}$ $3.7 \pm 0.1 \ ^{bA}$	$3.6 \pm 0.1 \ ^{aA}$ $3.6 \pm 0.2 \ ^{aAB}$	$3.3 \pm 0.1 \ ^{ m aB}$ $3.3 \pm 0.1 \ ^{ m aB}$
	Fresh-cut	C-CON C-TREAT	$\begin{array}{c} 3.3\pm0.1~^{\mathrm{xB}}\\ 3.4\pm0.1~^{\mathrm{xB}}\end{array}$	$\begin{array}{c} 3.8\pm0.2 ^{\text{xA}} \\ 3.6\pm0.1 ^{\text{xB}} \end{array}$	$3.8 \pm 0.1 \ ^{ m xB}$ $4.1 \pm 0.3 \ ^{ m xA}$	$\begin{array}{c} 3.4\pm0.1 ^{\text{xA}} \\ 3.4\pm0.1 ^{\text{xB}} \end{array}$
Total soluble solids (TSS, %)	Whole	W-CON W-TREAT	6.3 ± 0.3 $^{\mathrm{aA}}$ 6.3 ± 0.1 $^{\mathrm{aA}}$	$5.7\pm0.1~^{ m aB}$ $5.2\pm0.3~^{ m aB}$	$5.7\pm0.1~^{\mathrm{aB}}$ $5.8\pm0.3~^{\mathrm{aAB}}$	$5.3 \pm 0.2 \ ^{ m aB} 6.4 \pm 0.1 \ ^{ m b^*A}$
	Fresh-cut	C-CON C-TREAT	$5.9 \pm 0.2 \ ^{ m xAB}$ $6.3 \pm 0.3 \ ^{ m xA}$	6.5 ± 0.3 ^{xA} 5.9 ± 0.8 ^{xA}	$5.9 \pm 0.3 \ ^{ m xAB}$ $6.0 \pm 0.8 \ ^{ m xA}$	$5.6 \pm 0.1 \ ^{\mathrm{xB}}$ $5.8 \pm 0.1 \ ^{\mathrm{x^*A}}$
Titratable acidity (TA, mg L ⁻¹)	Whole	W-CON W-TREAT	8.7 ± 0.3 $^{\mathrm{aA}}$ 8.6 ± 0.2 $^{\mathrm{aA}}$	8.9 ± 0.8 ^{aA} 7.3 ± 0.2 ^{bB}	$6.6 \pm 0.4~^{ m aB}$ $7.0 \pm 0.6~^{ m aBC}$	$6.7\pm0.4~^{\mathrm{aB}}$ $6.2\pm0.3~^{\mathrm{aC}}$
	Fresh-cut	C-CON C-TREAT	9.5 ± 0.4 ^{xA} 9.0 ± 0.4 ^{xA}	$6.8 \pm 0.2 \ ^{ m xB}$ $7.0 \pm 0.8 \ ^{ m xB}$	$6.4 \pm 0.1 \ ^{ m xB} \ 6.6 \pm 0.2 \ ^{ m xB}$	6.2 ± 0.4 ^{xB} 6.1 ± 0.2 ^{xB}
Firmness (N)	Whole	W-CON W-TREAT	$1.7\pm0.2~^{\mathrm{aB}}$ $1.9\pm0.2~^{\mathrm{aA}}$	$2.2 \pm 0.3 \ ^{\mathrm{aB}}$ $1.6 \pm 0.3 \ ^{\mathrm{b^*A}}$	$1.9 \pm 0.2 \; ^{ m aB} 1.9 \pm 0.4 \; ^{ m aA}$	$2.9 \pm 0.2 \ ^{ m aA}$ $1.8 \pm 0.2 \ ^{ m aA}$
	Fresh-cut	C-CON C-TREAT	$\begin{array}{c} 2.4\pm0.3 \ ^{\mathrm{xA}} \\ 2.0\pm0.1 \ ^{\mathrm{yA}} \end{array}$	$1.7 \pm 0.2 \ ^{yA}$ $2.3 \pm 0.3 \ ^{x^*A}$	2.1 ± 0.6 ^{xA} 2.3 ± 0.8 ^{xA}	$2.5 \pm 1.1 \ ^{ m xA}$ $2.1 \pm 0.2 \ ^{ m xA}$
DPPH∙ (g kg ⁻¹)	Whole	W-CON W-TREAT	$\begin{array}{c} 4.907 \pm 0.610 \; ^{aA} \\ 5.482 \pm 0.247 \; ^{aA} \end{array}$	$\begin{array}{c} 5.664 \pm 0.251 \; ^{aA} \\ 5.613 \pm 0.321 \; ^{a^*A} \end{array}$	$\begin{array}{c} 5.471 \pm 0.224 \; ^{aA} \\ 5.566 \pm 0.287 \; ^{a^*A} \end{array}$	$\begin{array}{c} 5.421 \pm 0.736 \; ^{aA} \\ 5.244 \pm 0.239 \; ^{aA} \end{array}$
	Fresh-cut	C-CON C-TREAT	$\begin{array}{c} 5.642 \pm 0.382 \ ^{xAB} \\ 5.816 \pm 0.194 \ ^{xA} \end{array}$	$\begin{array}{c} 5.757 \pm 0.431 \ ^{yA} \\ 4.703 \pm 0.270 \ ^{y^*B} \end{array}$	$\begin{array}{c} 4.864 \pm 0.487 \text{ xAB} \\ 4.006 \pm 0.369 \text{ y*C} \end{array}$	$\begin{array}{c} 4.618 \pm 0.451 \ ^{\text{xB}} \\ 4.853 \pm 0.118 \ ^{\text{xB}} \end{array}$
FRAP (g kg ⁻¹)	Whole	W-CON W-TREAT	$\begin{array}{c} 1.366 \pm 0.106 \; ^{aA} \\ 1.323 \pm 0.018 \; ^{aA} \end{array}$	$\begin{array}{c} 1.455 \pm 0.062 \; ^{aA} \\ 1.436 \pm 0.034 \; ^{aA} \end{array}$	$\begin{array}{c} 1.434 \pm 0.027 \; ^{aA} \\ 1.480 \pm 0.044 \; ^{aA} \end{array}$	$\begin{array}{c} 1.391 \pm 0.088 \; ^{\rm aA} \\ 1.277 \pm 0.211 \; ^{\rm aA} \end{array}$
	Fresh-cut	C-CON C-TREAT	$\begin{array}{c} 1.305 \pm 0.020 \ ^{\text{xB}} \\ 1.373 \pm 0.049 \ ^{\text{xA}} \end{array}$	$\begin{array}{c} 1.492 \pm 0.057 \ ^{xA} \\ 1.269 \pm 0.011 \ ^{y^*B} \end{array}$	$\begin{array}{c} 1.288 \pm 0.091 \ ^{\text{xB}} \\ 1.179 \pm 0.033 \ ^{\text{y*C}} \end{array}$	$\begin{array}{c} 1.326 \pm 0.058 \ ^{\text{xB}} \\ 1.333 \pm 0.022 \ ^{\text{xAB}} \end{array}$
TPC (g kg ⁻¹)	Whole	W-CON W-TREAT	$\begin{array}{c} 0.229 \pm 0.013 \; ^{aA} \\ 0.226 \pm 0.006 \; ^{aA} \end{array}$	$\begin{array}{c} 0.233 \pm 0.009 \; ^{aA} \\ 0.230 \pm 0.007 \; ^{a^*\!A} \end{array}$	$\begin{array}{c} 0.234 \pm 0.013 \; ^{aA} \\ 0.210 \pm 0.028 \; ^{aA} \end{array}$	$\begin{array}{c} 0.221 \pm 0.004 \; ^{aA} \\ 0.205 \pm 0.025 \; ^{aA} \end{array}$
	Fresh-cut	C-CON C-TREAT		$\begin{array}{c} 0.227 \pm 0.016 \text{ xA} \\ 0.202 \pm 0.002 \text{ y}^{*\text{B}} \end{array}$		$\begin{array}{c} 0.208 \pm 0.005 \text{ xAB} \\ 0.210 \pm 0.003 \text{ xA} \end{array}$

	Format	Treatment ¹	D0	D4	D7	D11
TAC	Whole	W-CON W-TREAT	$\begin{array}{c} 0.028 \pm 0.001 \; ^{aA} \\ 0.027 \pm 0.001 \; ^{aA} \end{array}$	$\begin{array}{c} 0.028 \pm 0.001 \; ^{bA} \\ 0.031 \pm 0.001 \; ^{a^*B} \end{array}$	$\begin{array}{c} 0.029 \pm 0.001 \; ^{bA} \\ 0.032 \pm 0.001 \; ^{a^*B} \end{array}$	$\begin{array}{c} 0.029 \pm 0.001 \; ^{aA} \\ 0.029 \pm 0.002 \; ^{aAB} \end{array}$
$(g kg^{-1})$	Fresh-cut	C-CON C-TREAT	$\begin{array}{c} 0.022 \pm 0.001 \ ^{xA} \\ 0.027 \pm 0.001 \ ^{yAB} \end{array}$	$\begin{array}{c} 0.027 \pm 0.001 \; ^{xB} \\ 0.028 \pm 0.001 \; ^{x^*AB} \end{array}$	$\begin{array}{ccc} 0.028\pm 0.001 \ ^{xB} & 0.\\ 3 & 0.029\pm 0.001 \ ^{x^*A} & 0. \end{array}$	$\begin{array}{c} 0.027 \pm 0.001 \ ^{\text{xB}} \\ 0.027 \pm 0.001 \ ^{\text{xB}} \end{array}$
TAA	Whole	W-CON W-TREAT	$\begin{array}{c} 0.267 \pm 0.001 \; ^{aB} \\ 0.278 \pm 0.002 \; ^{aAB} \end{array}$	$\begin{array}{c} 0.282 \pm 0.001 \; ^{aA} \\ 0.288 \pm 0.003 \; ^{aA} \end{array}$		$\begin{array}{c} 0.219 \pm 0.001 \ ^{aC} \\ 0.227 \pm 0.006 \ ^{aB} \end{array}$
$(g kg^{-1})$	Fresh-cut	C-CON C-TREAT	$\begin{array}{c} 0.280 \pm 0.001 \ ^{\rm xA} \\ 0.276 \pm 0.001 \ ^{\rm xA} \end{array}$	$\begin{array}{c} 0.258 \pm 0.001 \ ^{\text{xB}} \\ 0.256 \pm 0.001 \ ^{\text{xA}} \end{array}$		$\begin{array}{c} 0.195 \pm 0.002 \; ^{\rm xD} \\ 0.233 \pm 0.002 \; ^{\rm yB} \end{array}$

Table 1. Cont.

¹ Treatments: W-CON, whole strawberries treated with control water; W-TREAT, whole strawberries treated with water-assisted ultraviolet-C with peracetic 40 mg L⁻¹; C-CON, fresh-cut strawberries treated with control NaOCl 200 mg L⁻¹; C-TREAT, fresh-cut strawberries treated with water-assisted ultraviolet-C with peracetic 40 mg L⁻¹.



Figure 2. Color, expressed as L*a*b* coordinates, in fresh and whole (**A**) or fresh and fresh-cut strawberries (**B**), after control (black lines) or WUVPA treatment (grey lines). (**C**) Color, expressed as L*a*b* coordinates, in frozen strawberries (-25 °C), with modified gas composition (black lines) or air composition (grey lines). Values are the mean of three repetitions ± standard deviation. Different letters show statistically significant differences (p < 0.05) among storage time (4 °C) within the same treatment (Tukey's HSD test), and asterisks show statistically significant differences (p < 0.05) between treatments on the same day.

Initial firmness values for whole fruits ranged from 2.4 to 1.7 N. In fresh-cut strawberries, statistical differences were observed between C-CON and C-TREAT. The firmness values of C-CON strawberries (2.4 ± 0.3 N) were significantly higher than those of C-TREAT strawberries (1.9 ± 0.1 N) (Table 1). The present study shows that firmness was maintained through 11 days of storage at 4 °C. In contrast, other studies reported a decrease in strawberries' firmness during storage after PA at 40 or 90 mg L^{-1} treatment [19,20]. Variations in study outcomes may be attributed to the maturity stage or cultivar of strawberries, which can impact firmness maintenance [21]. A study by Nunes et al. [21] concluded that mature strawberries (not yet ripe or overripe) retained their firmness during cold storage.

Regarding weight loss, all strawberries lost an average value of $2.9 \pm 0.2\%$ of their weight in the form of water, which was accumulated as drops in the internal surface of the package at the end of the storage period. This value was reached by the W-CON samples at D4, while other samples reached it at D11. Other studies have reported weight losses in whole strawberries packaged with an ozone atmosphere of 1.5% after 3 days of storage at 2 °C [22] or in whole strawberries packaged with ascorbic acid of 8 to 15% after 12 days of storage at 1 °C [23].

3.2.2. Antioxidant Capacity

The antioxidant capacity of the strawberries prior to any treatment was 5.462 ± 0.394 and 1.342 ± 0.033 g kg⁻¹ determined by DPPH· or FRAP methods, respectively (Table 1). The antioxidant values obtained by the FRAP method aligned with those from our previous studies on strawberries washed with 40 mg L⁻¹ PA [12]. However, values obtained via the DPPH· method were four times higher, possibly due to variations in the phenolic compound profile (including anthocyanins). These compounds, along with tannins, are recognized as the primary antioxidants in strawberries [24], employing different antioxidant mechanisms such as hydrogen atom or single electron transfer [25]. On the other hand, no differences were observed between controls and treatments. Antioxidant values obtained by FRAP were maintained at 1.332 ± 0.046 g kg⁻¹ at the end of storage, whereas by the DPPH· method, values had decreased by up to 4.735 ± 0.166 g kg⁻¹ in fresh-cut samples by the end of the storage period.

3.2.3. Phytochemical Changes

In this study, TPC and TAC values (Table 1) did not differ between formats immediately after the treatments or during storage. Despite the operational biosynthetic pathway for anthocyanins and phenolic compounds after harvest, storage at low temperatures is not expected to inhibit this process. The anthocyanin content was lower than is reported in the literature [26,27]. These differences could be attributed to fruit differences regarding maturity stage [28] or cultivar [9] or to different extraction methods [29].

The initial content of total ascorbic acid (TAA) in strawberries ranged from 0.267 to 0.280 g kg⁻¹. Results showing the principal changes in TAA during storage (Table 1) indicated that the application of WUVPA treatment was not detrimental for strawberry vitamin C content. The evolution of this vitamin in WUVPA samples was similar to that of their respective controls. Despite this, a sharper decrease in TAA was observed for the control half-cut strawberries (C-CON) when compared to whole fruits (W-CON, W-TREAT) and half-cut strawberries treated with WUVPA (C-TREAT). Generally, fruits exhibit a gradual degradation of vitamin C content as storage temperature or duration increases due to enhanced oxidation, which produces compounds devoid of vitamin function [30]. This degradation process can be delayed by low temperatures [31]. At the end of the storage period, the TAA of whole (W-CON and W-TREAT) and fresh-cut strawberries (C-TREAT) had decreased by 17.7%, while TAA content decreased by 29.1% in control fresh-cut strawberries (C-CON). Other authors have reported higher decreases (up to 50%) in strawberry vitamin C after 6 days of storage at 6 °C. Exposure to oxygen during storage can accelerate the oxidation of vitamin C [32].

3.2.4. Effect on Native Microbiota

The initial populations in strawberries are those obtained during W-CON treatment at D0, as these represented initial non-treated fruits. The initial TAM and Y&M counts were $2.3 \pm 0.1 \log \text{CFU g}^{-1}$ for both parameters (Table 2). The consistent evolution of TAM and Y&M observed during storage suggests that the native microbiota in strawberries is primar-

ily composed of yeasts and molds, as indicted by Ortiz-Solà et al. [33]. After treatments, Y&M counts were slightly reduced in W-TREAT, C-CON, and C-TREAT samples. This can be explained by the germicidal effect of UV-C light, which basically causes damage to the DNA structure [34]. PA is a highly effective biocide; it is a powerful oxidizer of C-C double bonds and reacts with sulfhydryl and sulfur bonds in proteins [35]. Reductions in bacterial pathogens were demonstrated by [10] in strawberries by using this combination (WUVPA). However, the decrease in the counts of natural occurring microbiota in strawberries found in that study was lower compared to that of artificially inoculated *L. monocytogenes* and *S. enterica* with the same sanitizing treatment [11]. De Sao Jose and Vanetti [19] reported a 3 log CFU g⁻¹ decrease in Y&M population after washing strawberries with 40 mg L⁻¹ PA. The higher reductions could be attributed to the treatment time (10 min), but that could be deleterious to strawberry firmness. Overall, during shelf life, populations were maintained at 3.1 log CFU g⁻¹. Even so, the EU regulation on microbial criteria for foodstuffs (EC 2073/2005 and subsequent modifications) does not include maximum levels of TAM in fresh and fresh-cut fruit.

Table 2. Total aerobic mesophylls (TAM) and yeast and mold (Y&M) counts (log CFU g⁻¹) in fresh strawberries. Values are the mean of three repetitions \pm standard deviation. Different lowercase letters show statistically significant differences (p < 0.05) between treatments within the same presentation (whole or fresh-cut): 'a' and 'b' are used to indicate differences between treatments within the whole fruit format, while 'x' and 'y' are used for the fresh-cut fruit (analyzed with contrast test). Asterisks show statistically significant differences (p < 0.05) between presentation formats (whole vs. fresh-cut) in samples treated with water-assisted ultraviolet-C with peracetic 40 ppm. Capital letters show statistically significant differences (p < 0.05) over time within the same treatment analyzed with Tukey's test.

	Format	Treatment ¹	D0	D4	D7	D11
TAM $(\log CFU g^{-1})$	Whole	W-CON W-TREAT	$2.3 \pm 0.1 \ ^{ m aB}$ $1.8 \pm 0.3 \ ^{ m aA}$	$2.7\pm0.2~^{\mathrm{aAB}}$ $1.9\pm0.4~^{\mathrm{aA}}$	2.9 ± 0.4 $^{\mathrm{aAB}}$ 1.0 ± 0.4 $^{\mathrm{bA}}$	$\begin{array}{c} 3.1\pm0.2~^{aA}\\ 1.0\pm0.1~^{b^*A} \end{array}$
	Fresh-cut	C-CON C-TREAT	$1.2 \pm 0.2 \ ^{\mathrm{xA}}$ $1.8 \pm 0.6 \ ^{\mathrm{xA}}$	1.0 ± 0.1 ^{xA} 1.7 ± 0.4 ^{xA}	$egin{array}{l} 1.0 \pm 0.1 \ ^{ m xA} \\ 1.0 \pm 0.1 \ ^{ m xA} \end{array}$	$egin{array}{ll} 1.0 \pm 0.1 \ ^{ m xA} \\ 1.6 \pm 0.1 \ ^{ m y^*A} \end{array}$
Y&M $(\log CFU g^{-1})$	Whole	W-CON W-TREAT	$2.3 \pm 0.4 \ ^{ m aB}$ $1.8 \pm 0.5 \ ^{ m aA}$	$2.5\pm0.1~^{\mathrm{aAB}}$ $2.1\pm0.8~^{\mathrm{aA}}$	2.9 ± 0.4 $^{\mathrm{aAB}}$ 1.3 ± 0.1 $^{\mathrm{bA}}$	3.0 ± 0.2 ^{aA} 1.9 ± 0.5 ^{bA}
	Fresh-cut	C-CON C-TREAT	$1.4 \pm 0.2 \ {}^{ m xA}$ $1.8 \pm 0.6 \ {}^{ m xA}$	1.4 ± 0.2 ^{xA} 1.8 ± 0.4 ^{xA}	$1.5 \pm 0.3 \ ^{ m xA}$ $1.3 \pm 0.1 \ ^{ m xA}$	$\begin{array}{c} 1.7 \pm 0.1 \ ^{\rm xA} \\ 1.6 \pm 0.2 \ ^{\rm xA} \end{array}$

¹ Treatments: W-CON, whole strawberries treated with control water; W-TREAT, whole strawberries treated with water-assisted ultraviolet-C with peracetic 40 mg L⁻¹; C-CON, fresh-cut strawberries treated with control NaOCl 200 mg L⁻¹; C-TREAT, fresh-cut strawberries treated with water-assisted ultraviolet-C with peracetic 40 mg L⁻¹.

3.3. Frozen Strawberries

3.3.1. Commercial Quality of the Fruits

The percentage of O_2 and CO_2 was maintained throughout all the storage, as expected in frozen products. One of the purposes of the MAP application was to protect strawberries from detrimental changes during thawing. However, no significant effect on quality or biochemical parameters was observed in thawed strawberries that could be attributed to air conditions inside the package.

Regarding quality parameters (pH, TSS, and TA), the results can be observed in Table 3. Although statistical analysis revealed that the differences between pH values before and after the washing treatments were statistically significant, the pH values measured moved within the reported range for strawberries [16]. The TSS content did not vary ($6.2 \pm 0.4\%$.), neither when strawberries were washed in WUV-C and PA 40 mg L⁻¹ nor after the frozen process. In contrast, TA showed a significant decrease in strawberries that were frozen (from 7.8 ± 0.3 to 6.6 ± 0.2 mg L⁻¹) but increased afterwards during the 12 months of frozen storage.

Table 3. Quality assessment of strawberries: pH, total soluble solids (TSS, %), titratable acidity (TA, mg L⁻¹), firmness, and biochemical parameters: changes in antioxidant activity, expressed by DPPH· and FRAP values (mg AAE/100 g FW); total phenolic content (TPC, mg GAE/100 g FW); total anthocyanin content (TAC, mg pelargonidin-3-glucosyde/100 g FW); and total ascorbic acid (mg AA/100 g FW) in frozen strawberries. Values are the mean of three repetitions \pm standard deviation. Different lowercase letters show statistically significant differences (p < 0.05) between gas compositions on the same day, and capital letters show statistically significant differences (p < 0.05) over time within the same treatment analyzed with Tukey's test. Underlined values show statistically significant differences (p < 0.05) between the three steps (initial, after WUVPA ¹, and frozen-thawed at M0), analyzed with Tukey's test.

	Packaging	Before WUVPA ¹	After WUVPA ¹	M0	М3	M6	M12
рН	MAP ² Air	3.7 ± 0.1	$\underline{3.5\pm0.1}$	$3.8 \pm 0.1 \ ^{aA}$ $3.6 \pm 0.1 \ ^{aA}$	$3.6 \pm 0.1 \ ^{\mathrm{aB}}$ $3.5 \pm 0.1 \ ^{\mathrm{aB}}$	$3.7 \pm 0.1 \ ^{aA}$ $3.7 \pm 0.1 \ ^{aA}$	3.8 ± 0.1 $^{\mathrm{aA}}$ 3.8 ± 0.1 $^{\mathrm{aA}}$
Total soluble solids (TSS, °Brix)	MAP ² Air	5.8 ± 0.3	6.4 ± 0.3	$6.3 \pm 0.3 \ ^{\mathrm{aA}}$ $5.9 \pm 0.5 \ ^{\mathrm{aA}}$	$5.9 \pm 0.5 \ ^{\mathrm{aA}}$ $6.0 \pm 0.5 \ ^{\mathrm{aA}}$	$6.3 \pm 0.3 ~^{\mathrm{aA}}$ $6.4 \pm 0.2 ~^{\mathrm{aA}}$	6.2 ± 0.4 $^{\mathrm{aA}}$ 7.0 ± 0.4 $^{\mathrm{aA}}$
Titratable acidity (TA, mg L ⁻¹)	MAP ² Air	7.8 ± 0.3	7.8 ± 0.4	$\frac{6.7 \pm 1.1}{6.5 \pm 0.8}^{\rm aB}$	$6.5 \pm 1.3 \; ^{ m aB} \ 6.6 \pm 1.0 \; ^{ m aB}$	$7.5\pm1.7~^{\mathrm{aAB}}$ $8.2\pm0.4~^{\mathrm{aAB}}$	$8.7 \pm 1.4 \; {}^{\mathrm{aA}}$ $7.9 \pm 0.3 \; {}^{\mathrm{aA}}$
Firmness (N)	MAP ² Air	1.6 ± 0.1	1.6 ± 0.1	$\frac{0.7\pm0.1}{0.8\pm0.1}^{\mathrm{aA}}_{\mathrm{aA}}$	$1.2\pm0.8~^{\mathrm{aA}}$ $1.1\pm0.5~^{\mathrm{aA}}$	1.1 ± 0.3 $^{\mathrm{aA}}$ 1.1 ± 0.3 $^{\mathrm{aA}}$	1.3 ± 0.2 aA 1.3 ± 0.1 aA
DPPH· $(g kg^{-1})$	MAP ² Air	6.948 ± 0.565	7.848 ± 0.610	$\begin{array}{l} 6.919 \pm 0.742 \; ^{\rm aA} \\ 7.594 \pm 0.078 \; ^{\rm aA} \end{array}$	$\begin{array}{c} 6.183 \pm 0.316 \ ^{\rm aAB} \\ 6.798 \pm 0.680 \ ^{\rm aAB} \end{array}$	$\begin{array}{l} 4.799 \pm 0.378 \; ^{\rm aC} \\ 4.845 \pm 0.641 \; ^{\rm aC} \end{array}$	$\begin{array}{l} 6.619 \pm 0.760 \; ^{aB} \\ 6.255 \pm 0.394 \; ^{aB} \end{array}$
FRAP (g kg ⁻¹)	MAP ² Air	7.594 ± 0.617	7.744 ± 0.544	$\begin{array}{l} 7.400 \pm 0.543 \; ^{aA} \\ 7.859 \pm 0.646 \; ^{aA} \end{array}$	$\begin{array}{l} 6.929 \pm 0.528 \ ^{aAB} \\ 8.048 \pm 0.288 \ ^{aAB} \end{array}$	$\begin{array}{l} 6.001 \pm 0.337 \; ^{aC} \\ 6.257 \pm 0.763 \; ^{aC} \end{array}$	$\begin{array}{l} 6.677 \pm 0.693 \; {}^{\mathrm{aBC}} \\ 6.687 \pm 0.522 \; {}^{\mathrm{aBC}} \end{array}$
$\begin{array}{c} \text{TPC} \\ (g \ kg^{-1}) \end{array}$	MAP ² Air	0.156 ± 0.012	0.177 ± 0.013	$\frac{0.424 \pm 0.029}{0.449 \pm 0.035} {}^{aA}$	$\begin{array}{c} 0.307 \pm 0.023 \ ^{aB} \\ 0.368 \pm 0.007 \ ^{bB} \end{array}$	$\begin{array}{c} 0.185 \pm 0.012 \; ^{aC} \\ 0.214 \pm 0.038 \; ^{aC} \end{array}$	$\begin{array}{c} 0.075 \pm 0.016 \; ^{aD} \\ 0.072 \pm 0.015 \; ^{aD} \end{array}$
$\begin{array}{c} \text{TAC} \\ (g \text{ kg}^{-1}) \end{array}$	MAP ² Air	0.025 ± 0.003	0.026 ± 0.020	$\begin{array}{c} 0.029 \pm 0.001 \; ^{aA} \\ 0.028 \pm 0.003 \; ^{aA} \end{array}$	$\begin{array}{c} 0.022 \pm 0.002 \; ^{aA} \\ 0.021 \pm 0.002 \; ^{aA} \end{array}$	$\begin{array}{c} 0.026 \pm 0.001 \; ^{aA} \\ 0.026 \pm 0.011 \; ^{aA} \end{array}$	$\begin{array}{c} 0.027 \pm 0.001 \ ^{aA} \\ 0.024 \pm 0.003 \ ^{aA} \end{array}$
${\rm TAA} \atop ({\rm g}{\rm kg}^{-1})$	MAP ² Air	0.306 ± 0.026	0.315 ± 0.020	$\begin{array}{c} 0.304 \pm 0.010 \; ^{aA} \\ 0.295 \pm 0.060 \; ^{aA} \end{array}$	$\begin{array}{c} 0.312 \pm 0.050 \; ^{aA} \\ 0.324 \pm 0.010 \; ^{aA} \end{array}$	$\begin{array}{c} 0.246 \pm 0.060 \ ^{aB} \\ 0.261 \pm 0.018 \ ^{aA} \end{array}$	$\begin{array}{c} 0.294 \pm 0.043 \; ^{aA} \\ 0.286 \pm 0.015 \; ^{aA} \end{array}$

 1 WUVPA, water-assisted ultraviolet-C light combined with peracetic acid at 40 mg L $^{-1}$. 2 MAP, modified atmosphere packaging.

The firmness of strawberries evaluated before and after the washing treatment did not vary and was maintained at 1.6 \pm 0.1 N (Table 3). However, firmness decreased after freezing and thawing, regardless of the storage time. Frozen fruits are especially susceptible to damage during the freezing process, resulting in the softening of texture, loss of water holding capacity, and deterioration of colors. It is accepted that this occurs with the formation of ice crystals, increasing the final volume and damaging the cell lamellae of fruits [36]. However, a quick freezing process may reduce this damage [36]. The measurement of drip loss revealed that after thawing, strawberries had lost between 4.7 ± 2.6 and $10.4 \pm 0.5\%$ of their weight in form of water, resulting in a turgor loss in the cell tissue. Drip loss is typically attributed to three main factors: high internal pressure in the product, the formation of ice crystals, and the irreversibility of water removal from cells [37]. On the other hand, these factors depend on the matrix and on the freezingthawing conditions [38]. For instance, Holzwarth, Korhummel, Carle, and Kammerer [39] evaluated the effects of various thawing conditions (4 and 37 °C) and durations (8 to 48 h) on frozen strawberries (with liquid N₂) stored at -20 °C, with drip losses of between 9.1 ± 0.0 and $19.2 \pm 0.4\%$.

The color of frozen strawberries remained unchanged after washing with WUVPA (Figure 2C). No changes were observed after freezing and thawing after 0 and 3 months of storage. However, after 6 and 12 months of storage, there was a decrease in L* and b* values, regardless of the atmosphere in the packaging. The color parameters at M12 were 35.8 ± 0.8 , 40.6 ± 1.4 , and 19.2 ± 1.4 for L*, a*, and b*, respectively, indicating a darker and

more purplish color. It is highly important to implement freezing and thawing methods that enable color retention. Other authors have also reported a darkening in the typical red color, regardless of whether a pretreatment using a pulsed electric field coupled with vacuum infusion of cryoprotectants was applied to frozen–thawed strawberries [40]. In contrast, Holzwarth, Korhummel, Carle, and Kammerer [39] observed that when strawberries were thawed at low temperatures and for short periods (4 °C and 8 h), the color parameters were better retained than when thawing was performed at higher temperatures (20 or 37 °C).

3.3.2. Antioxidant Capacity

The initial values of DPPH· and FRAP were 6.948 ± 0.565 and 7.594 ± 0.617 g kg⁻¹, respectively (Table 3). These values were maintained immediately after washing with WUVPA. Although some authors have noted a decline in the antioxidant capacity of strawberries upon freezing [41], a significant decrease in antioxidant capacity was observed only after 6 months of storage in the present study. Antioxidant formation in strawberries is a delicate system that can be easily affected. According to the literature, it can be attributed to phenolic compounds, anthocyanins, and vitamin C [24,42].

3.3.3. Phytochemical Changes

The initial TPC value was 0.156 ± 0.012 g kg⁻¹ (Table 3). After freezing, the value increased significantly to 0.424-0.449 g kg⁻¹. This increase in TPC value could be explained by the collapse of the cell wall caused by the formation of frozen crystals, and the subsequent liberation of some phenolic compounds that were not previously accessible for their determination [36]. However, the TPC value decreased gradually with storage time (after 12 months at $-20 \,^{\circ}$ C, it was 0.072-0.075 g kg⁻¹). Exposure to oxygen during storage can promote oxidation reactions, reducing the TPC [32].

The initial TAC values were 0.025 and 0.026 g kg⁻¹, before and after WUVPA treatment, respectively. The content of anthocyanins was maintained throughout the 12 months of storage, similar to what other authors have reported. Anthocyanins are relatively stable compounds compared to other phytochemicals present in strawberries. They are less susceptible to degradation caused by enzymatic activity, oxidation, or fluctuations in temperature and humidity [43].

In the present study, the TAA content ranged from 0.306 ± 0.026 to 0.294 ± 0.043 g kg⁻¹ for strawberries prior to any treatment and for strawberries that had undergone 12 months of storage, respectively (Table 3). Similarly, other authors have reported the maintenance of initial ascorbic acid values in frozen strawberries over time [44,45]. Vitamin C is very labile, so if a given process keeps its levels relatively unchanged, it is likely that most other nutrients have survived the process as well [44].

3.3.4. Effect on Native Microbiota

The initial TAM and Y&M counts were both $2.5 \pm 0.1 \log \text{CFU g}^{-1}$. After washing treatment, the counts decreased by 0.5 and 0.8 log units for TAM and Y&M, respectively. Regardless of the storage time, there were no significant differences between the beginning and the end of the shelf life of the frozen strawberries (from 0 to 12 months), since the TAM and Y&M counts after freezing remained at 1.4 ± 0.1 and $1.3 \pm 0.1 \log \text{CFU g}^{-1}$, respectively (see Table S1 in Supplementary Material). The microbial populations in frozen strawberries probably decreased due to the cell damage that occurred during freezing. Although the formation of intracellular ice should be lower in quick freezing than in conventional freezing, the formation of intracellular ice affecting microbial viability could also occur.

4. Conclusions

The present study showed that combining peracetic acid and ultraviolet-C light effectively extended the shelf life of both fresh and fresh-cut strawberries. This sanitation method proved to be comparable to chlorine washing for maintaining fruit quality. Despite a slight decrease in firmness after freezing, the nutritional content of the strawberries remained stable during long-term storage.

Supplementary Materials: The following supporting information can be downloaded at: https:// www.mdpi.com/article/10.3390/horticulturae10050452/s1, Table S1. Total aerobic mesophylls (TAM) and yeasts and molds (Y&M) counts (log CFU g⁻¹) in frozen strawberries. Values are the mean of 3 repetitions \pm standard deviation. Different lowercase letters show statistically significant differences (p < 0.05) between gas compositions in the same day, and capital letters show statistically significant differences (p < 0.05) during time within the same treatment analyzed with Tukey's test. Underlined values show statistically significant differences (p < 0.05) between the three steps (initial, after WUVPA, and frozen–thawed at M0), analyzed with Tukey's test; Table S2. Graphical representation of the strawberries (whole and fresh-cut) after the respective treatment over the days; Table S3. Graphical representation of the strawberries (frozen) after their respective treatment; Table S4. Gas composition (O₂ and CO₂) throughout the shelf-life of whole and cut strawberries; Table S5. Average respiration rate (RR) of different strawberry formats (whole and fresh-cut) througout the shelf-life (11 d); Table S6. Weight loss of the different strawberry formats (whole and cut) throughout the shelf-life of the product.

Author Contributions: I.N.-L.: data curation, formal analysis, methodology, writing original and edited draft. J.O.: data curation, formal analysis, methodology, writing original draft. I.V.: project administration, funding acquisition. M.A.: methodology, supervision, writing review. G.B.: methodology, validation, visualization. I.A.-A.: project administration, supervision, writing review and editing. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by 'Ministerio de Educación, Economía y Cultura' for the financial support of the project AGL2016-78086-R. This work was also supported by the CERCA Programme of Generalitat de Catalunya. I. Nicolau-Lapeña is in receipt of a predoctoral grant awarded by the 'Ministerio Español de Economía, Industria y Competitivdad' (grant number BES-2017-079779). J. Ortiz-Solà is in receipt of a predoctoral grant awarded by Universitat de Lleida (grant number (BOU186-243/2017 UdL). Dr. I. Aguiló-Aguayo thanks the National Programme for the Promotion of Talent and Its Employability of the 'Ministerio de Economía, Industria y Competitividad' of the Spanish Government and the European Social Fund for her Postdoctoral Senior Grant 'Ramon y Cajal' (RYC-2016-2019 949). The authors also would like to thank S. Villaró, J. Costa, and M. Anguera for their technical support. The authors would like to thank Carburos Metalicos S.A. for kindly providing the freezing tank.

Data Availability Statement: The data presented in this study are available on request from the corresponding author.

Conflicts of Interest: The authors declare no conflicts of interests.

Abbreviations

Plate count agar (PCA); dichloran rose bengale chloramphenicol agar (DRBC); 2,4,6-tris(2pyridyl)-s-triazine (TPTZ); 2,2-diphenyl-1-picrylhydrazyl (DPPH); 3,3',3''-phosphanetriyltripropanoic acid (TCEP); water-assisted UV-C (WUV-C); peracetic acid (PA); water-assisted UV-C with peracetic acid (WUVPA); oxidation-reduction potential (ORP); millivolts (mV); turbidity units (NTU); whole strawberry control (W-CON); whole strawberry treated (W-TREAT); fresh-cut control (C-CON); fresh-cut treated (C-TREAT); sodium hypochlorite (NaOCl); polypropylene (PP); nitrogen (N₂); modified atmosphere packaging (MAP); respiration rate (RR); ferric reducing antioxidant power (FRAP); total phenolic content (TPC); total anthocyanin content (TAC); total ascorbic acid (TAA); high-performance liquid chromatography (HPLC); total aerobic mesophylls (TAM); yeasts and molds (Y&M); colony-forming unit (CFU); honest significant difference (HSD); total soluble solids (TSS); titratable acidity (TA).

References

1. Gani, A.A.; Baba, W.N.; Ahmad, M.; Shah, U.; Khan, A.A.; Wani, I.A.; Masoodi, F.A.; Gani, A.A. Effect of Ultrasound Treatment on Physico-Chemical, Nutraceutical and Microbial Quality of Strawberry. *LWT-Food Sci. Technol.* **2016**, *66*, 496–502. [CrossRef]

- 2. Pinzon, M.I.; Sanchez, L.T.; Garcia, O.R.; Gutierrez, R.; Luna, J.C.; Villa, C.C. Increasing Shelf Life of Strawberries (*Fragaria* ssp.) by Using a Banana Starch-Chitosan-*Aloe vera* Gel Composite Edible Coating. *Int. J. Food Sci. Technol.* **2019**, *55*, 92–98. [CrossRef]
- Hazarika, T.K.K.; Lalrinfeli, L.; Lalchhanmawia, J.; Mandal, D. Alteration of Quality Attributes and Shelf-Life in Strawberry (*Fragaria × ananassa*) Fruits during Storage as Influenced by Edible Coatings. *Indian J. Agric. Sci.* 2019, 89, 28–34. [CrossRef]
- 4. Kelly, K.; Madden, R.; Emond, J.P.; do Nascimento Nunes, M.C. A Novel Approach to Determine the Impact Level of Each Step along the Supply Chain on Strawberry Quality. *Postharvest Biol. Technol.* **2019**, *147*, 78–88. [CrossRef]
- 5. Yang, Z.; Cao, S.; Cai, Y.; Zheng, Y. Combination of Salicylic Acid and Ultrasound to Control Postharvest Blue Mold Caused by *Penicillium expansum* in Peach Fruit. *Innov. Food Sci. Emerg. Technol.* **2011**, *12*, 310–314. [CrossRef]
- 6. Bovi, G.G.; Rux, G.; Caleb, O.J.; Herppich, W.B.; Linke, M.; Rauh, C.; Mahajan, P.V. Measurement and Modelling of Transpiration Losses in Packaged and Unpackaged Strawberries. *Biosyst. Eng.* **2018**, *174*, 1–9. [CrossRef]
- Luo, Y.; Zhou, B.; Van Haute, S.; Nou, X.; Zhang, B.; Teng, Z.; Turner, E.R.; Wang, Q.; Millner, P.D. Association between Bacterial Survival and Free Chlorine Concentration during Commercial Fresh-Cut Produce Wash Operation. *Food Microbiol.* 2018, 70, 120–128. [CrossRef] [PubMed]
- 8. European Food Safety Authority. Scientific Opinion on the Risk Posed by Pathogens in Food of Non-Animal Origin. Part 2 (*Salmonella* and Norovirus in Berries). *EFSA J.* **2014**, *12*, 3832. [CrossRef]
- Šamec, D.; Maretić, M.; Lugarić, I.; Mešić, A.; Salopek-Sondi, B.; Duralija, B. Assessment of the Differences in the Physical, Chemical and Phytochemical Properties of Four Strawberry Cultivars Using Principal Component Analysis. *Food Chem.* 2016, 194, 828–834. [CrossRef] [PubMed]
- Nicolau-Lapeña, I.; Abadias, M.; Viñas, I.; Bobo, G.; Lafarga, T.; Ribas-Agustí, A.; Aguiló-Aguayo, I. Water UV-C Treatment Alone or in Combination with Peracetic Acid: A Technology to Maintain Safety and Quality of Strawberries. *Int. J. Food Microbiol.* 2020, 335, 108887. [CrossRef]
- Ortiz-Solà, J.; Valero, A.; Abadias, M.; Nicolau-Lapeña, I.; Viñas, I. Evaluation of Water-Assisted UV-C Light and its Additive Effect with Peracetic Acid for the Inactivation of *Listeria monocytogenes*, *Salmonella enterica* and Murine Norovirus on Whole and Fresh-Cut Strawberries during Shelf-Life. J. Sci. Food Agric. 2022, 102, 5660–5669. [CrossRef] [PubMed]
- 12. Nicolau-Lapeña, I.; Abadias, M.; Bobo, G.; Aguiló-Aguayo, I.; Lafarga, T.; Viñas, I. Strawberry Sanitization by Peracetic Acid Washing and Its Effect on Fruit Quality. *Food Microbiol.* **2019**, *83*, 159–166. [CrossRef] [PubMed]
- Meyers, K.J.; Watkins, C.B.; Pritts, M.P.; Liu, R.H. Antioxidant and Antiproliferative Activities of Strawberries. J. Agric. Food Chem. 2003, 51, 6887–6892. [CrossRef] [PubMed]
- 14. Lafarga, T.; Bobo, G.; Viñas, I.; Zudaire, L.; Simó, J.; Aguiló-Aguayo, I. Steaming and Sous-Vide: Effects on Antioxidant Activity, Vitamin C, and Total Phenolic Content of Brassica Vegetables. *Int. J. Gastron. Food Sci.* **2018**, *13*, 134–139. [CrossRef]
- 15. Montanez, J.C.; Rodríguez, F.A.S.; Mahajan, P.V.; Frías, J.M. Modelling the Effect of Gas Composition on the Gas Exchange Rate in Perforation-Mediated Modified Atmosphere Packaging. *J. Food Eng.* **2010**, *96*, 348–355. [CrossRef]
- 16. Costa, F.B.; Duarte, P.S.; Puschmann, R.; Finger, F.L. Quality of Fresh-Cut Strawberry. Hortic. Bras. 2011, 29, 477–484. [CrossRef]
- 17. Aday, M.S.; Temizkan, R.; Büyükcan, M.B.; Caner, C. An Innovative Technique for Extending Shelf Life of Strawberry: Ultrasound. *LWT-Food Sci. Technol.* **2013**, *52*, 93–101. [CrossRef]
- 18. Aday, M.S.; Caner, C. Individual and Combined Effects of Ultrasound, Ozone and Chlorine Dioxide on Strawberry Storage Life; Elsevier Ltd.: Amsterdam, The Netherlands, 2014; Volume 57. [CrossRef]
- 19. de São José, J.F.B.; Vanetti, M.C.D. Application of Ultrasound and Chemical Sanitizers to Watercress, Parsley and Strawberry: Microbiological and Physicochemical Quality. *LWT-Food Sci. Technol.* **2015**, *63*, 946–952. [CrossRef]
- 20. Guo, M.; Jin, T.Z.; Gurteler, J.B.; Fan, X.; Yadav, M.P. Inactivation of *Escherichia coli* O157:H7 and *Salmonella* and Native Microbiota on Fresh Strawberries by Antimicrobial Washing and Coating. *J. Food Prot.* **2018**, *81*, 1227–1235. [CrossRef] [PubMed]
- Nunes, M.C.N.; Morais, A.M.M.B.; Brecht, J.K.; Sargent, S.A. Fruit Maturity and Storage Temperature Influence Response of Strawberries to Controlled Atmospheres. J. Am. Soc. Hortic. Sci. 2002, 127, 836–842. [CrossRef]
- Nadas, A.; Olmo, M.; García, J.M. Growth of Botrytis Cinerea and Strawberry Quality in Ozone-Enriched Atmospheres. *Food Microbiol. Saf.* 2003, 68, 1798–1802. [CrossRef]
- 23. Sogvar, O.B.O.B.; Koushesh Saba, M.; Emamifar, A.; Saba, M.K.; Emamifar, A. *Aloe vera* and Ascorbic Acid Coatings Maintain Postharvest Quality and Reduce Microbial Load of Strawberry Fruit. *Postharvest Biol. Technol.* **2016**, *114*, 29–35. [CrossRef]
- Fierascu, R.C.; Temocico, G.; Fierascu, I.; Ortan, A.; Babeanu, N.E. *Fragaria* Genus: Chemical Composition and Biological Activities. *Molecules* 2020, 25, 498. [CrossRef] [PubMed]
- Schlesier, K.; Harwat, M.; Böhm, V.; Bitsch, R. Assessment of Antioxidant Activity by Using Different In Vitro Methods. *Free Radic. Res.* 2002, *36*, 177–187. [CrossRef] [PubMed]
- Van De Velde, F.; Vaccari, M.C.; Piagentini, A.M.; Pirovani, M.É. Optimization of Strawberry Disinfection by Fogging of a Mixture of Peracetic Acid and Hydrogen Peroxide Based on Microbial Reduction, Color and Phytochemicals Retention. *Food Sci. Technol. Int.* 2016, 22, 485–495. [CrossRef] [PubMed]
- 27. Li, M.; Li, X.; Han, C.; Ji, N.; Jin, P.; Zheng, Y. UV-C Treatment Maintains Quality and Enhances Antioxidant Capacity of Fresh-Cut Strawberries. *Postharvest Biol. Technol.* 2019, 156, 110945. [CrossRef]
- 28. Ban, Z.; Zhang, J.; Li, L.; Luo, Z.; Chen, C.; Wang, Y.; Yuan, Q.; Cai, C.; Yu, L. Antioxidant Profiles of Strawberry (*Fragaria* × *ananassa*) in Relation to Fruit Maturity and Postharvest Storage. J. Biobased Mater. Bioenergy **2018**, 12, 122–128. [CrossRef]

- Azmir, J.; Zaidul, I.S.M.; Rahman, M.M.; Sharif, K.M.; Mohamed, A.; Sahena, F.; Jahurul, M.H.A.; Ghafoor, K.; Norulaini, N.A.N.; Omar, A.K.M. Techniques for Extraction of Bioactive Compounds from Plant Materials: A Review. J. Food Eng. 2013, 117, 426–436. [CrossRef]
- 30. Adisa, V.A. The Influence of Molds and Some Storage Conditions in Ascorbic Acid Content in Orange and Pineapple Fruits. *Food Chem.* **1986**, *21*, 139–146. [CrossRef]
- 31. Lee, S.K.; Kader, A.A. Preharvest and Postharvest Factors Influencing Vitamin C Content of Horticultural Crops. *Postharvest Biol. Technol.* **2000**, *20*, 207–220. [CrossRef]
- 32. Cordenunsi, B.R.; Nascimento, J.R.O.; Lajolo, F.M. Physico-Chemical Changes Related to Quality of Five Strawberry Fruit Cultivars during Cool-Storage. *Food Chem.* **2003**, *83*, 167–173. [CrossRef]
- Ortiz-Solà, J.; Viñas, I.; Colás-Medà, P.; Anguera, M.; Abadias, M. Occurrence of Selected Viral and Bacterial Pathogens and Microbiological Quality of Fresh and Frozen Strawberries Sold in Spain. *Int. J. Food Microbiol.* 2020, 314, 108392. [CrossRef] [PubMed]
- Barba, F.J.; Koubaa, M.; do Prado-Silva, L.; Orlien, V.; Sant'Ana, A.d.S. Mild Processing Applied to the Inactivation of the Main Foodborne Bacterial Pathogens: A Review. *Trends Food Sci. Technol.* 2017, 66, 20–35. [CrossRef]
- 35. Rossi, S.; Antonelli, M.; Mezzanotte, V.; Nurizzo, C. Peracetic Acid Disinfection: A Feasible Alternative to Wastewater Chlorination. *Water Environ. Res.* 2007, *79*, 341. [CrossRef]
- 36. Kobayashi, R.; Suzuki, T. Effect of Supercooling Accompanying the Freezing Process on Ice Crystals and the Quality of Frozen Strawberry Tissue. *Int. J. Refrig.* **2019**, *99*, 94–100. [CrossRef]
- 37. Jul, M. Quality Frozen Foods, 1st ed.; Academic Press: London, UK, 1984; pp. 377–415.
- Sirijariyawat, A.; Charoenrein, S. Freezing Characteristics and Texture Variation after Freezing and Thawing of Four Fruit Types. Songklanakarin J. Sci. Technol. 2012, 34, 517–523.
- Holzwarth, M.; Korhummel, S.; Carle, R.; Kammerer, D.R. Evaluation of the Effects of Different Freezing and Thawing Methods on Color, Polyphenol and Ascorbic Acid Retention in Strawberries (*Fragaria × ananassa* Duch.). *Food Res. Int.* 2012, 48, 241–248. [CrossRef]
- 40. Velickova, E.; Tylewicz, U.; Dalla, M.; Winkelhausen, E.; Kuzmanova, S.; Romani, S. Effect of Pulsed Electric Field Coupled with Vacuum Infusion on Quality Parameters of Frozen/Thawed Strawberries. *J. Food Eng.* **2018**, 233, 57–64. [CrossRef]
- Marques, K.K.; Renfroe, M.H.; Brevard, P.B.B.; Robert, E.; Gloeckner, J.W.; Marques, K.K.; Renfroe, M.H.; Brevard, P.B.B.; Robert, E. Differences in Antioxidant Levels of Fresh, Frozen and Freeze-Dried Strawberries and Strawberry Jam. *Int. J. Food Sci. Nutr.* 2010, *61*, 759–769. [CrossRef]
- 42. Alvarez-Suarez, J.M.; Mazzoni, L.; Forbes-Hernandez, T.Y.; Gasparrini, M.; Sabbadini, S.; Giampieri, F. The Effects of Pre-Harvest and Post-Harvest Factors on the Nutritional Quality of Strawberry Fruits: A Review. J. Berry Res. 2014, 4, 1–10. [CrossRef]
- Oliveira, A.; Coelho, M.; Alexandre, E.M.C.; Almeida, D.P.F. Long-Term Frozen Storage and Pasteurization Effects on Strawberry Polyphenols Content. *Food Bioprocess Technol.* 2015, *8*, 1838–1844. [CrossRef]
- 44. Bouzari, A.; Holstege, D.M.; Barrett, D.M. Vitamin Retention in Eight Fruits and Vegetables: A Comparison of Refrigerated and Frozen Storage. *J. Agric. Food Chem.* **2015**, *63*, 957–962. [CrossRef] [PubMed]
- 45. Li, L.; Pegg, R.B.; Eitenmiller, R.R.; Chun, J.; Kerrihard, A.L. Ected Nutrient Analyses of Fresh, Fresh-Stored, and Frozen Fruits and Vegetables. J. Food Compos. Anal. 2020, 59, 8–17. [CrossRef]

Disclaimer/Publisher's Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.