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The Impact of Post-Harvest Treatment on the Preservation of "San Andreas" Strawberries

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Abstract

Strawberries have bioactive and organoleptic properties; however, their mechanical and pathological sensitivity is a challenge to the large-scale marketing of this fruit. Techniques such as sanitization and rapid pre-storage cooling are applied to extend the shelf life and market distribution of strawberries. The objective of this study was to evaluate the efficacy of pre-and post-harvest sanitization methods on the preservation of "San Andreas" strawberries. The methods employed included the application of peracetic acid and hydrocooling, calcium chloride treatments, and a combination of heat treatment and post-harvest sanitizer. The strawberries were stored in polyethylene terephthalate containers at room temperature and in a cold room. Regardless of storage conditions, the combination of hydrocooling and peracetic acid significantly reduced mass loss and maintained fruit firmness. This treatment extended the strawberries' shelf life by up to nine days compared to the control group ($p < 0.05$). In addition to assisting in the maintenance of the fruit's epidermis, refrigerated storage or the application of hydrocooling helped to preserve the physicochemical and bioactive properties of the strawberry.

Keywords

Hydrocooling, Peracetic Acid, *Fragaria x ananassa*, Shelf-Life

1. Introduction

Strawberries (*Fragaria x ananassa*) are cultivated extensively in Brazil using cultivars that overcome edaphoclimatic challenges and enhance production potential [1]. Among the factors that influence plant growth, photoperiod plays a crucial role in their metabolism and phenological development. The vegetative stage benefits from increased temperature and day length, while shorter days and lower

temperatures lead to floral induction. The day-neutral cultivars San Andreas and Aromas, are used for cultivation in the southern region of Brazil, as they have requirements compatible with the climatic conditions and good physical and chemical quality for commercial purposes, and also the "San Andreas" cultivar is known for its high productivity and resistance to diseases such as gray mold (*Botrytis cinerea*) and anthracnose (*Colletotrichum acutatum*), which are essential traits for ensuring high yields and reducing the need for pesticides [2]-[4]. The variety was also donated by a producer from Ponta Grossa, who already uses it successfully, ensuring reliability in its selection and validation for the region.

Strawberry production in Brazil is still limited, especially in the foreign market [5]. This is due to the fruit's sensitivity to mechanical and pathological damage, which can compromise visual quality during commercialization [6]. Given that it is a non-climacteric fruit, strawberries show few metabolic changes following harvest, with a reduction in respiration rate and senescence [7].

The use of chemical control is frequently employed in strawberry production due to the high susceptibility of the fruit to pathogens during the various stages of ripeness. Pathogens of the genera Botrytis and Rhizopus are especially damaging when the fruit is approaching physiological ripeness and can cause significant losses in fruit production and storage [8]. Chemical, thermal, or sanitizing treatments can be applied to reduce the development of pathogens. Peracetic acid (an organic oxide with rapid biocidal action and no residue) and calcium (which strengthens the cell wall by forming pectates that are resistant to fungal enzymes) are examples of such treatments [9].

Physical control methods, such as forced cooling and hydrocooling, reduce respiration and fruit senescence, thereby extending the post-harvest life of the fruit [10]. It is common practice in Brazil to refrigerate strawberries at 0˚C with high humidity (95%) to preserve them for up to seven days. This method of refrigeration prevents degradation and maintains organoleptic quality [6]. Hydrocooling, which entails immersing or spraying the fruit with ice water, provides rapid and uniform cooling without causing physical damage [11]. The method can reduce microbial activity without compromising the integrity of the fruit when applied at a temperature of 2˚C [12].

The two treatments complement each other synergistically, while hydro-cooling slows down physiological deterioration, peracetic acid prevents the proliferation of microorganisms. This combined strategy can result in an extended shelf life for strawberries compared to conventional methods, without compromising food safety or the sensory quality of the fruit. Additionally, it addresses the growing demand for more sustainable post-harvest methods by reducing the use of synthetic fungicides and minimizing chemical residues [13]. Furthermore, studies on other vegetables, such as lettuce and green onions, indicate that hydro-cooling is effective in reducing mass loss and maintaining visual quality. Hydro-cooling at 4˚C for 5 minutes, followed by storage at 5˚C, helps maintain the water balance in the leaves, delaying wilting and extending the product's shelf life [14] [15]. Alt-

hough these studies do not directly involve strawberries, the results suggest that hydro-cooling may be a promising technique for prolonging the shelf life of various horticultural products.

The use of resistant packaging (such as polyethylene plastic tubs with PVC film) is essential to reduce mechanical damage and make commercialization easier. However, this packaging can lead to the development of a microclimate that favors the proliferation of pathogens [16]. The objective of this study was to evaluate the efficacy of pre-and post-harvest sanitization methods on the preservation of "San Andreas" strawberries. The data in this study was previously used in a master's thesis [17] which evaluated the effectiveness of pre-and-post-harvest sanitization and post-harvest hydrocooling, and extend the shelf life of "San Andreas" strawberries.

2. Materials and Methods

Strawberries of the San Andreas cultivar produced in the city of Ponta Grossa, Paraná, in a protected cultivation system with fertigation in slabs without prior phytosanitary management were used. In addition to the control, five different treatments were analyzed: pre-harvest application of peracetic acid; a combination of pre-harvest peracetic acid and post-harvest hydrocooling; post-harvest hydrocooling only; hydrocooling followed by post-harvest application of $CaCl_2$; and a combination of post-harvest hydrocooling and peracetic acid.

The greenhouse area was divided into two sections, with only one receiving a single dose of PRO15® on the strawberry plants. The PRO15® was dissolved in a ratio of 15 ml per 10 L of water, following the instructions provided by the manufacturer. The compound has a sanitizing action through peracetic acid and is recommended for use on strawberry crops, with no deficiency or residual action.

The fruits were harvested when at least two-thirds of their epidermis exhibited pigmentation. They were later transported to the Laboratory of Biotechnology Applied to Fruit Farming at the State University of Ponta Grossa (UEPG). The strawberries were then divided into those that had or had not received a pre-harvest application of peracetic acid for subsequent post-harvest treatment. A total of 10 L of distilled water were used, with 1/4 of the solution in liquid form and 3/4 frozen, until the solution temperature was adjusted to 1˚C. The hydro-cooling process consisted of immersing 20 kg of fruit into the solution, maintaining a 1:2 water-to-fruit ratio. The fruits being monitored and kept in the solution until they achieved 7/8 of the total cooling [18]. This was observed after one hour of immersion, when the pulp temperature reached 2.5˚C. The treatment involving the addition of calcium chloride ($CaCl_2$) utilized the hydro-cooled medium with 2% soluble $CaCl_2$. For the post-harvest peracetic acid treatment, 15 ml of the product was added to the hydrocooled medium. The fruits were randomly selected and packed in PET containers with ventilation holes to facilitate airflow and maintain humidity. The packages had a mass of approximately 80 g and were stored at room temperature (20˚C ± 2˚C) and in a cold room (0˚C ± 2˚C) without relative hu-

midity control.

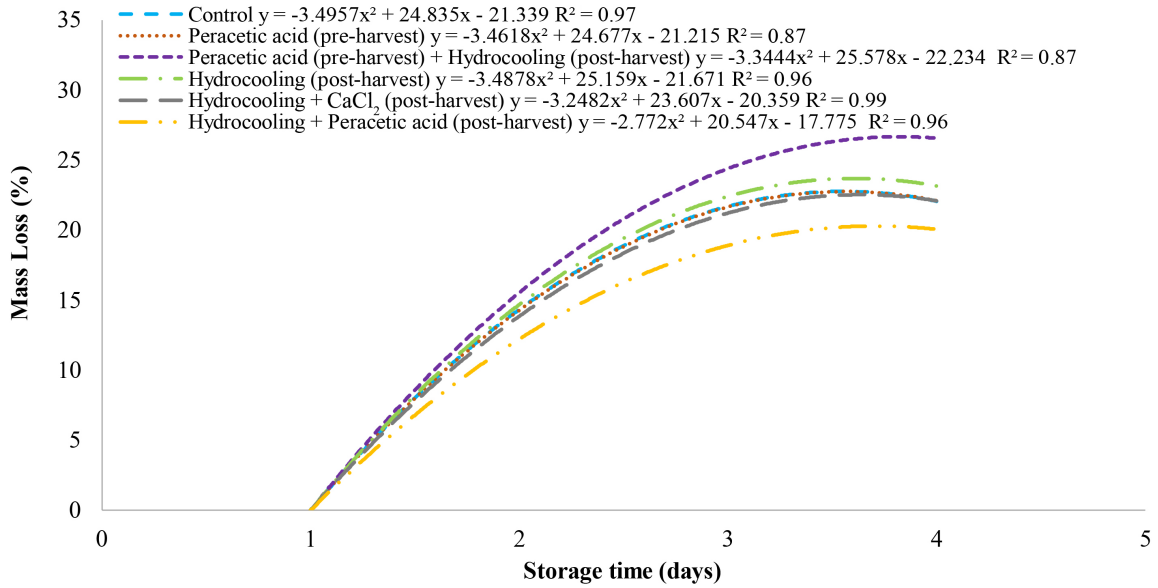
The physicochemical analyses were carried out one day after harvest. The fruits at room temperature were evaluated after 2 and 4 days while the fruits in cold storage were evaluated after 7 and 9 days in cold storage, plus 2 days at room temperature, simulating commercial settings. The five most homogeneous strawberries in each package were subjected to analysis to determine the loss of mass, firmness, total soluble solids (SS), titratable acidity, converted into % citric acid, anthocyanin content, analyzed by the method described by Lee and Francis and total polyphenols, analyzed according to the method described by Singleton and Rossi which uses the Folin-Ciocalteau reagent, both adapted by Nunes, Brecht, Morais and Sargent [19].

The experiment was conducted in a completely randomized design (CRD), considering the homogeneity of the units, with four replications for each treatment. Statistical analyses were carried out using the software SISVAR 5.6, the data was organized for an initial evaluation and then subjected to regression analysis and the F-test ($p < 0.05$), analysis of variance (ANOVA) with the comparison of means using the Tukey test ($p < 0.05$) and SNK ($p < 0.05$) for non-normal data.

3. Results and Discussion

3.1. Mass Loss

The treatments were evaluated for up to four days, given that this was the maximum period in which the fruit remained in conditions suitable for consumption at room temperature. The regression analysis indicated a statistical difference at 5% between the different storage periods for all treatments, showing a proneness to lose mass (Graph 1).



Graph 1. Mass loss (%) of "San Andreas" strawberries after 1, 2, and 4 days of storage at room temperature. Source: The authors.

The decrease in fresh mass is due to transpiration, which results in the loss of water through the tissues. This process compromises the quality and sale ability of the fruit. Lack of or delayed post-harvest cooling is a crucial factor in maintaining transpiration [20] [21]. In 2022, Anami *et al.* observed that delayed cooling increases the vapor pressure between the strawberry and the air, resulting in a significant loss of water in the tissues [22]. The oxidative action of peracetic acid might have caused damage to the membrane by releasing active oxygen, which degrades the structure of proteins and enzymes in the plasma membrane. When associated with the low temperature from hydrocooling, the transpiration rate can be intensified [23]. When comparing the mass loss of fruit in each treatment, it was observed that the hydrocooling + peracetic acid treatment (post-harvest) showed the lowest loss at the end of storage (20.06%).

The maximum mass loss for strawberries in the post-harvest phase without compromising the quality of the fruit for fresh consumption is between 6% and 10% [24] [25]. Also, mass loss is one of the main indicators of post-harvest quality, and losses above 10% can negatively affect consumer acceptance [26]. This may have occurred due to the drying out of the epidermis, which prevents the permeation of water through the tissues (**Figure 1**) [6].

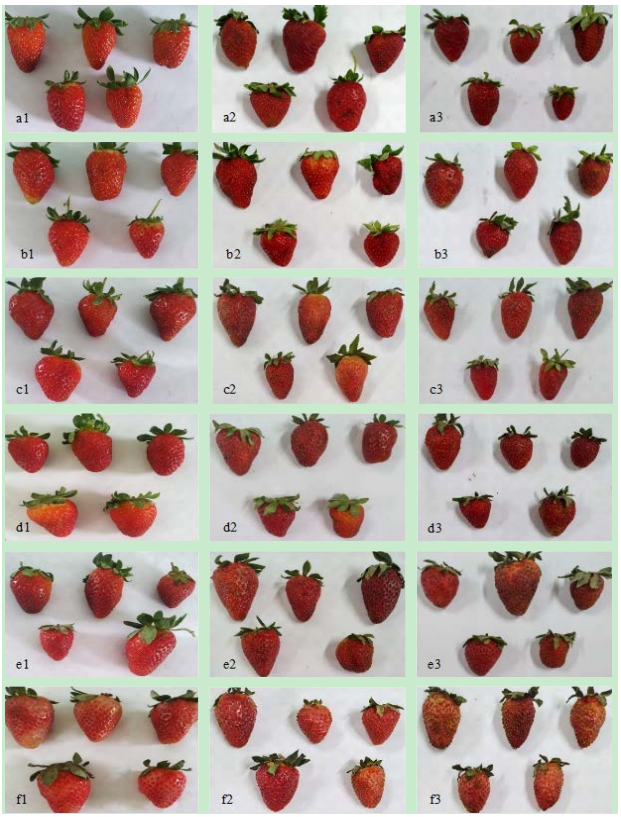


Figure 1. "San Andreas" strawberries after 1, 2 and 4 days of storage at room temperature. a = Control; b = Peracetic acid (pre-harvest); c = Peracetic acid (pre-harvest) + Hydrocooling (post-harvest); d = Hydrocooling (post-harvest); e = Hydrocooling + $CaCl_2$ (post-harvest); f = Hydrocooling + Peracetic acid (post-harvest). 1 = 1 day; 2 = 2 days; 3 = 3 days after storage at 20˚C. Source: The authors.

Although the observed mass loss exceeded this range, *Nunes et al.* reported a 15% loss in "Sweet Charlie" strawberries after delayed cooling) [20]. Hydrocooling was effective in containing mass loss, and when combined with peracetic acid (post-harvest) showed the lowest loss in both cooling periods (14.33% and 18.28%), compared to the control (19.80% and 23.48%) (Table 1).

Table 1. Mass Loss (%) of "San Andreas" Strawberries after 1, 7, and 9 days of storage in a cold chamber followed by 2 days at room temperature.

Treatments	Storage time (days)	
	7 + 2	9 + 2
Control	19.80 Ba	23.48 Aa
Peracetic acid (pre-harvest)	16.47 Bab	19.88 Aab
Hydrocooling (post-harvest)	15.66 Bab	18.21 Ab
Hydrocooling + Peracetic acid (post-harvest)	14.33 Bb	18.28 Ab
CV (%)	18.18	18.63
Means	16.60 B	19.96 A

Means with same uppercase letter in the row and lowercase letter in the column are not significantly different by the SNK test ($p < 0.05$). Source: The authors.

The literature shows that the internal temperature of strawberries increases due to cellular respiration even after cooling. This increase in temperature results in a loss of mass and water, caused by thermal and water stress [21]. The literature shows that the internal temperature of strawberries increases due to cellular respiration even after cooling. This increase in temperature results in a loss of mass and water, caused by thermal and water stress [27].

3.2. Firmness

Firmness is directly related to the cells' turgor capacity and tissue resistance [28]. The treatments did not differ throughout storage at room temperature or refrigeration (Table 2), likewise, *Ferreira et al.* observed no variation in firmness after subjecting "Sweet Charlie" strawberries to hydrocooling and storing them in a cold chamber [27].

Table 2. Firmness (N) of "San Andreas" strawberries after 1, 2, and 4 days of storage at room temperature.

Treatments	Storage time (days)			Means
	1	2	4	
Control	13.22 a	14.40 a	12.07 a	13.23 a
Peracetic acid (pre-harvest)	11.45 a	12.62 a	13.02 a	12.36 a
Peracetic acid (pre-harvest) + Hydrocooling (post-harvest)	12.60 a	10.72 a	9.27 a	10.86 a

Continued

Hydrocooling (post-harvest)	13.87 a	11.25 a	14.05 a	13.05 a
Hydrocooling + $CaCl_2$ (post-harvest)	13.70 a	13.95 a	10.35 a	12.66 a
Hydrocooling + Peracetic acid (post-harvest)	10.95 a	9.82 a	13.90 a	11.55 a
CV (%)	15.40	16.80	27.00	
Means	12.63	12.12	12.11	

Means with same letter in column are not significantly different by the Tukey's test ($p < 0.05$). Source: The authors.

Were reported a high firmness that is characteristic of the San Andreas cultivar, which may have influenced the lower loss of firmness during storage [29] [30]. In Figure 2, it can be noted that the strawberries treated with hydrocooling and peracetic acid (post-harvest) showed drying of the epidermis, a factor that may have contributed to the higher firmness.

Figure 2. "San Andreas" after 7 and 9 days of storage in a cold chamber (0˚C ± 2˚C) + 2 days at room temperature (20˚C). a = Control; b = Peracetic acid (pre-harvest); c = Hydrocooling (post-harvest); d = Hydrocooling + Peracetic acid (post-harvest). 1 = 7 + 2 days; 2 = 9 days at 0˚C + 2 days after storage at 20˚C. Source: The authors.

The extreme post-harvest handling of strawberries should be avoided, as it affects the external structure due to the high sensitivity of the tissues, especially when exposed to high temperatures [31]. The stage of ripeness, storage temperature, and the action of enzymes such as polygalacturonase (PG) influence the quality of the fruit tissues, potentially degrading the walls and causing cell leakage due to the greater permeability of the membranes in the senescence process, which reduces firmness and increases the susceptibility of the fruit to mechanical damage [32]. Mechanical damage in strawberries is identified by the loss of turgidity. As a result, fresh mass and firmness are reduced, affecting visual freshness and the brightness of the skin [29]. In 2006, Nunes studied three varieties of strawberries and observed that the less pigmented the epidermis, the higher the firmness of the fruit and that when kept in cold storage the firmness of the fruit gradually decreased due to the natural progression of ripening [19]. Despite this, he discovered that fruit with 75% of the epidermis pigmented under refrigeration may not show a reduction in the firmness of the flesh, as they are in the final stages of ripening.

3.3. °Brix

There was a reduction in soluble solids at room temperature for the hydrocooling and hydrocooling + peracetic acid (post-harvest) treatments (7.26 and 6.66), with the lowest statistical average (6.46 °Bx) being observed in the fruit from the hydrocooling (post-harvest) treatment (**Table 3**).

Table 3. Soluble solids (SS expressed in °Brix) of "San Andreas" strawberries after 1, 2, and 4 days of storage at room temperature.

Treatments	Storage time (days)			Means
	1	2	4	
Control	6.53 Bb	7.71 Aa	7.52 Ab	7.25 b
Peracetic acid (pre-harvest)	8.03 ABa	7.85 Ba	8.70 Aa	8.19 a
Peracetic acid (pre-harvest) + Hydrocooling (post-harvest)	7.27 Aab	7.02 Aa	7.26 Abc	7.8 b
Hydrocooling (post-harvest)	7.04 Aab	7.06 Aa	6.46 Ac	6.85 b
Hydrocooling + $CaCl_2$ (post-harvest)	6.71 Bb	6.97 Ba	7.47 Abc	7.05 b
Hydrocooling + Peracetic acid (post-harvest)	7.10 Aab	6.93 Aa	6.66 Abc	6.92 b
CV (%)	7.62	5.77	7.02	
Means	7.13 A	7.26 A	7.35 A	

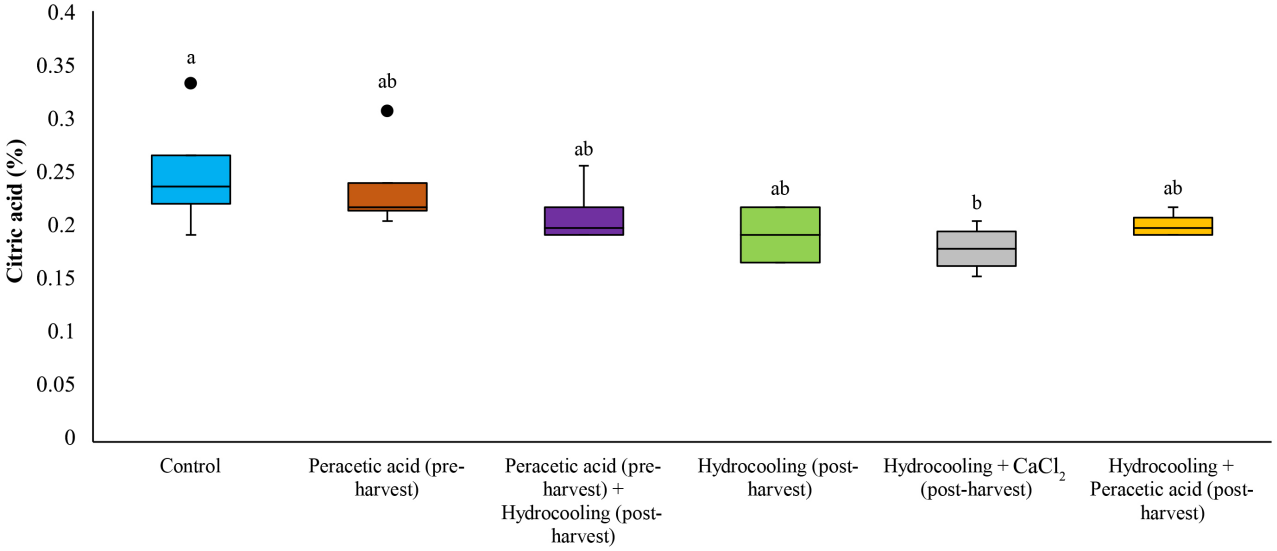
Means with same uppercase letter in the row and lowercase letter in the column are not significantly different by the Tukey's test ($p < 0.05$). Source: The authors.

In general, there is a tendency for the sugar content to rise throughout storage. It was observed that peracetic acid (pre-harvest) can significantly influence the accumulation of total sugars since it showed the highest average (8.70) after 4 days

of storage. This condition may be related to its ability to influence permeability and membrane degradation, causing an accumulation of soluble solids [17]. There was no statistical difference in the results for fruit that remained in cold storage, as observed by *Jacomino et al.* in 2011 who evaluated the application of hydrocooling in "Festival" strawberries [11].

3.4. Citric Acid (%)

The highest acidity value was observed after 1 day in the fruit treated with peracetic acid (pre-harvest) + hydrocooling (post-harvest) (0.22%), the control had the highest average citric acid (0.25%) after 2 days of ambient storage; the only treatment that differed from this was the hydrocooling + $CaCl_2$ (0.18%) with the lowest average among the others (**Graph 2**).

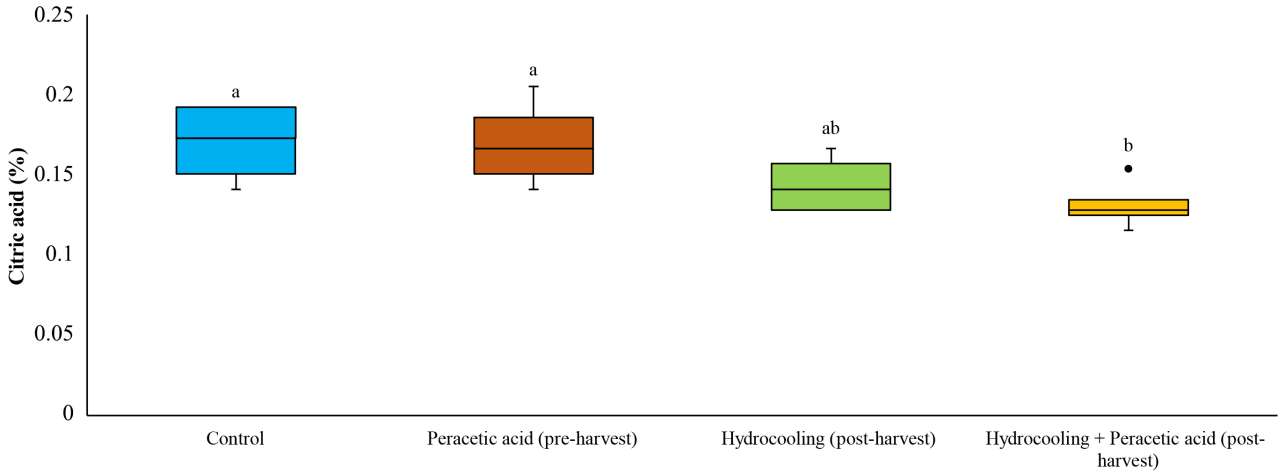


Graph 2. Citric acid (%) of "San Andreas" strawberries after 2 days of storage at room temperature. Treatment means followed by the same lowercase letter do not differ by SNK test ($p < 0.05$). Source: The authors.

A review of the literature reveals comparable findings, indicating that hydrocooling does not act internally, therefore preventing the degradation of organic acids during storage [33]. The fruit kept under refrigeration did not show significantly different results from the control (0.13), however, the storage period had an influence on the fruit from the hydrocooling + peracetic acid treatment (post-harvest), showing 0.15% after one day, 0.20% and 0.13% after seven and nine days refrigerated + two at room temperature, respectively (**Graph 3**).

On the last day of storage, this treatment was responsible for the lowest average acidity compared to the other treatments and the control, which had the highest (0.17%). Following refrigerated storage, an increase in citric acid concentration was observed. However, some authors posit that this phenomenon is contingent upon storage conditions since the utilization of hydrocooling as a preservation method has not been demonstrated to correlate with the maintenance of acidity

in strawberries. These shoes that despite being non-climacteric, there are chemical variations during post-harvest due to the senescence process, with a gradual and constant reduction in respiratory rate. After reaching physiological maturity, there is a decrease in the production and supply of exogenous ethylene, one of the main phytohormones responsible for maintaining fruit maturity. Although ethylene levels were not monitored in this study, it is known that post-harvest, this hormone causes tissue degradation, which can be minimized with life extension techniques [34] [35]. This condition can be mitigated with the application of shelf-life prolongation techniques because, despite the respiration rate in non-climacteric fruit after harvest, the synthesis and action of ethylene are continuous and can compromise the integrity of the tissues [35].

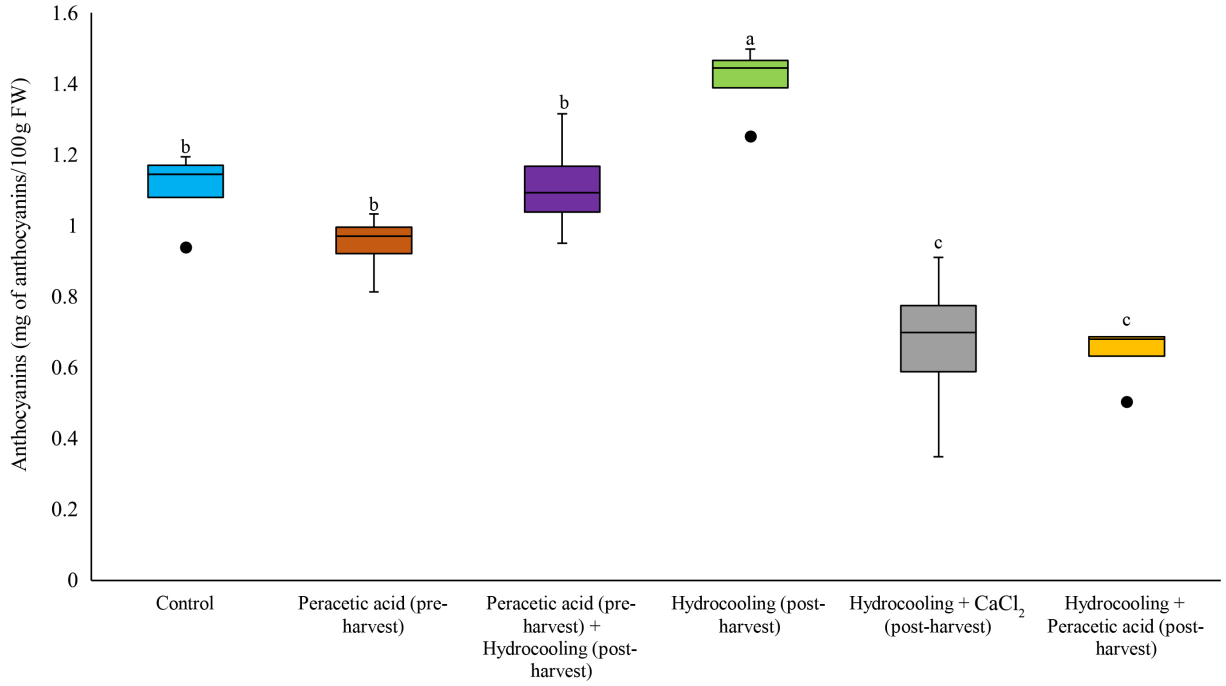


Graph 3. Citric acid (%) of "San Andreas" strawberries after 9 days of storage in a cold chamber followed by 2 days at room temperature. Treatment means followed by the same lowercase letter do not differ by SNK test ($p < 0.05$). Source: The authors.

3.5. Anthocyanins

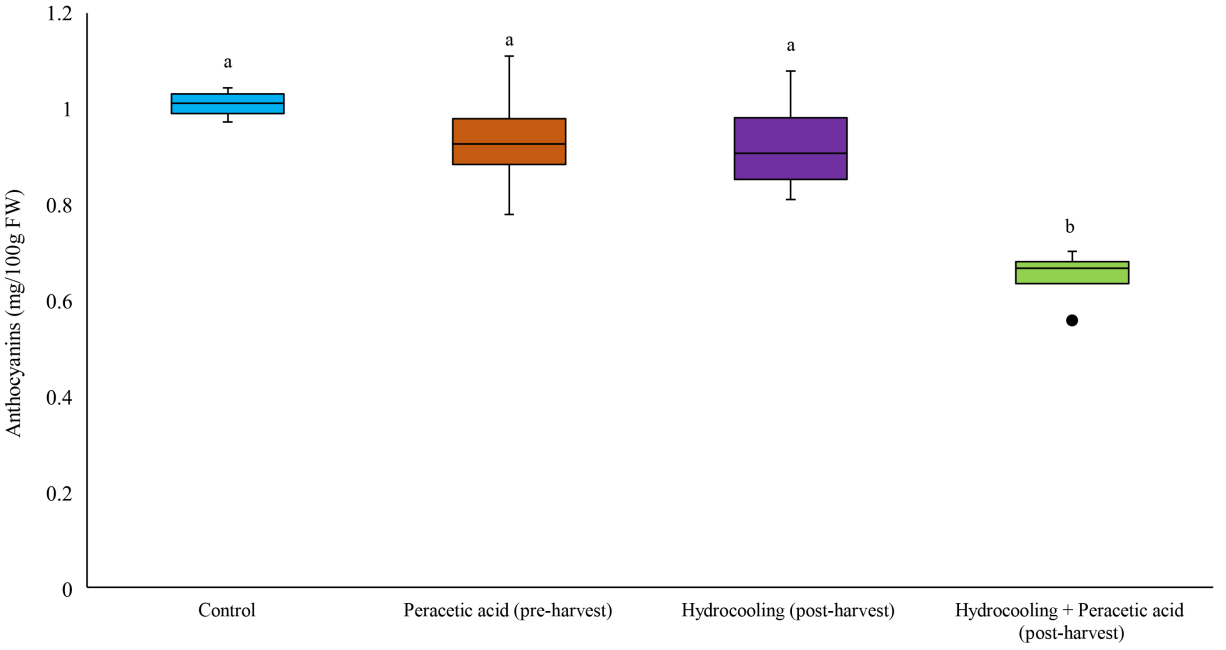
Hydrocooling (post-harvest) had the highest overall average (1.081 mg of anthocyanins/100g FW (fresh weight)) compared to the other treatments. It is observed that the fruit subjected to hydrocooling differed from the control, in which it is possible to see that when the fruits are isolated, they have the highest average (1.409 mg) and when associated with some sanitizing agent, they have the lowest (0.663 and 0.638 mg) (**Graph 4**).

Keeping the fruit in ambient condition for two days was the limit for maintaining high concentrations of anthocyanins without external darkening due to pigment degradation. Anthocyanins continue to accumulate even after harvest, particularly at elevated temperatures [19]. Both thermal and sanitizing interactions demonstrated a decline in saturation (**Figure 1**). Furthermore, the anthocyanin content may be influenced if there is any damage to the epidermis or exposure to any molecule, as this will result in the pigments diffusing through the tissues, rendering the fruit opaque and darkened. In 2019, *Nicolau-Lapena et al.* observed that the longer the period of exposure of strawberries to peracetic acid, the higher the accumulation of pigments [36].



Graph 4. Anthocyanins (mg/100g FW) of "San Andreas" strawberries after 2 days of storage at room temperature. Treatment means followed by the same lowercase letter do not differ by SNK test ($p < 0.05$). Source: The authors.

In this study, the treatments differed from each other after 9 days in refrigeration + 2 days at room temperature, with the fruit with hydrocooling (post-harvest) displaying the lowest average compared to the others (0.649 mg) (**Graph 5**).



Graph 5. Anthocyanins (mg/100g FW) of "San Andreas" strawberries after 9 days of storage in a cold chamber followed by 2 days at room temperature. Treatment means followed by the same lowercase letter do not differ by Tukey's test ($p < 0.05$). Source: The authors.

Recent studies have demonstrated that refrigeration effectively minimizes oxidative stress in strawberries by reducing the production of reactive oxygen species (ROS), that helps preserve the integrity of anthocyanins, the primary flavonoid pigments responsible for the fruit's red color [37] [38]. Additionally, maintaining refrigerated storage conditions supports the activity of antioxidant enzymes such as superoxide dismutase (SOD) and catalase (CAT), which play crucial roles in mitigating oxidative damage and preserving the epidermal color of the fruit [39].

Carnelossi et al., in 2019 found no significant differences in hydrocooled versus untreated blueberries during refrigerated storage, but a reduction in anthocyanin content occurred, which was linked to enzymatic pigment degradation [40]. This degradation may be associated with fruit senescence, which degrades chemical compounds, particularly bioactive molecules. Alterations in epidermal structure also influence anthocyanin levels, as mechanical damage leads to pigment diffusion through internal tissues, resulting in external opacity [41].

3.6. Polyphenols

There was a statistical difference by regression analysis in the peracetic acid (pre-harvest) ($y = 3.4577x^2 + 5.4278x + 791.18$, R2 = 0.90), hydrocooling (post-harvest) ($y = -13.811x^2 + 82.2x + 722.99$, R2 = 0.55) and hydrocooling + peracetic acid (post-harvest) ($y = -23.319x^2 + 127.25x + 688.06$, R2 = 0.52) treatments, as they showed a variation in the total phenolics content (TPC) between storage periods. The hydrocooling + peracetic acid treatment (post-harvest) showed an increase in TPC with the highest average after two days (849.28 mg) and a subsequent reduction. On the other hand, the peracetic acid (pre-harvest) and hydrocooling (post-harvest) treatments showed a continuous increase in TPC with the highest averages after four days at room temperature (868.22 and 845.30 mg). There was no difference between the treatments applied regarding ambient and refrigerated storage and the storage period by the Tukey test at 5%. The overall average total polyphenol content was 849.12 mg and 858.17 mg after seven and nine days of refrigerated storage + two days at room temperature. Just as observed for anthocyanins, it was possible to determine that the storage condition does not promote the preservation of phenolic compounds. Were observed that there can be a reduction of more than 20% in the total content of phenolics in strawberries and no significant difference with the application of sanitizing agents and subsequent refrigerated storage [36] [42]. The production of phenolic compounds and their derivatives increases in stressful situations; therefore, thermal assets and sanitizers can affect the concentrations of this compounds [17].

4. Conclusion

Hydrocooling, both alone and combined with peracetic acid, increased the shelf life of strawberries by up to four days at room temperature. This is due to the supply of water and rapid cooling after harvest, which minimizes transpiration losses. In cold room conditions, the fruit lasted seven days plus two additional

days at room temperature, as this method helps to delay senescence and reduce the occurrence of pathogens. Although the treatment causes changes in the epidermis due to the sensitivity of the strawberries, it does not affect the bioactive composition or cause surface darkening.

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Conflicts of Interest

All authors declare that there is no conflict of interest in the authorship of this paper.

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