Egypt. J. Chem. Vol. 67, SI: M. R. Mahran, pp. **2089 - 2100** (2024)

Egyptian Journal of Chemistry

http://ejchem.journals.ekb.eg/

Biodegradable Nanoparticle Coating from Chitosan, Salicylic Acid and Banana Peel Extract Enhances Shelf Life of "Murcott " Fruits

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Abstract

Murcott "mandarins are a promising export variety due to their appealing appearance and quality. However, improper postharvest handling and storage led to 30-50% loss of the crop. This study investigated the use of biodegradable edible coatings to improve the quality and storage life of Murcott mandarins. Edible coatings; prepared from banana peel extracts (BPE) 10% and 20%, chitosan nanoparticles (CHNPs) 2% and 3%, and salicylic acid nanoparticles (SANPs) 0.25% and 0.5%; were applied on the fruit for 5 minutes. Coatings were applied individually or in combination, and the fruits were stored for 100 days at 5±1°C and 90±1% relative humidity. Shelf-life evaluation was also conducted for 6 days at room temperature. Fruit decay, weight loss, total loss, color, firmness, total soluble solids (TSS), total acidity, ascorbic acid content, respiration rate, and sensory quality were assessed every 20 days. BPE, CHNPs, and SANPs had positive effects on the physiochemical characteristics, storage capacity, and overall quality of Murcott mandarins. Dipping fruits in a solution of BPE (20%), CHNPs (3%), and SANPs (0.5%) for 5 minutes reduced decay, weight loss, and total loss. It also helped maintain fruit firmness, color, acidity, and ascorbic acid content while minimizing respiration rate throughout storage and higher levels of SOD, POD, and PPO activities. Sensory quality was also maintained. In conclusion, the combined use of BPE (20%), CHNPs (3%), and SANPs (0.5%) for 5 minutes significantly enhanced the fruit quality and storage potential of Murcott mandarins under cold storage conditions.

Keywords: *Citrus reticulata* L. ; storability; eco-friendly; respiration rate; antioxidant enzymes activity.

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1. Introduction

The Citrus fruits, such as mandarins, are known for their abundant antioxidant compounds, such as flavonoids, hydroxycinnamic acid, and vitamin C. These compounds contribute to their strong antioxidant activity[1]. In Egypt, the Murcott variety is widely cultivated, but it faces challenges in terms of losses during and after harvest, which ultimately reduce its postharvest lifespan[2]. Compared to other citrus fruits, mandarin varieties [3], in particular, tend to lose their quality at a faster rate between harvest and consumption due to their higher water content and nutrient composition[4]. Postharvest losses can be significant, ranging from 30 to 50% of the overall crop, mainly attributed to fruit metabolic disorders and fungal diseases[1]. To counter these issues, citrus fruits often undergo various treatments during export preparation, such as disinfection, immersion in chemical fungicide solutions, and waxing with fungicide-infused wax. These measures aim to reduce microbial load and protect the fruit from infections, thus minimizing losses. However, with the increasing demand for safer and healthier food, the food industry has been actively researching alternative methods to preserve fruits and vegetables without relying heavily on chemical fungicides[5,6].

Agricultural waste, including banana peels, is typically disposed of in landfills or used as agricultural fertilizer. The amount of waste generated during banana preparation is considerable, with the peel alone accounting for 35-40% of the fresh fruit's weight [7,8]. The peel of bananas, an often overlooked byproduct, contains insoluble nutritional fibers such as pectin, lignin, cellulose, and hemicellulose[9]. From a green chemistry perspective, the composition of banana peels offers various possibilities for creating sustainable technologies that reduce the environmental impact caused by discarded agro-food waste and associated costs. These include the development of food coating films, cellulose nanofibers, pectin products, ethanol, and phenolic compounds[10–12].

The potential of using banana peel as a promising material for edible coatings has been demonstrated in postharvest applications, particularly on "Prata" bananas. It has been found that treating the bananas with an 80% ethanol BPE resulted in the best retention of color, flavor, and texture[13]. Additionally, [14] discovered that coating apples with BPE improved fruit quality after harvest[15].

Chitosan (CH), one of the most abundant natural polymers, can be derived from algae, fungi cell walls,

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Receive Date: 06 August 2024, Revise Date: 11 September 2024, Accept Date: 10 October 2024
DOI: 10.21608/ejchem.2024.310412.10152

crustaceans' exoskeletons, and insect cuticles[16]. It is considered an excellent and safe edible coating for various fruit varieties[17]. CH and CHNPs have shown success in reducing postharvest deterioration and extending the storage period for mandarins, "Valencia " orange [18,19], apricots[20], and guavas[21].

Salicylic acid (SA), a naturally occurring plant growth regulator, inhibits ethylene synthesis and delays fruit senescence. It is also a safe method for controlling decay and preserving fruit quality after harvest[22,23]. The use of SA has been shown to enhance storability by reducing respiration rate, delaying color change and softening, preserving sugars, acids, and aroma, preventing chilling injury, stimulating pathogen resistance, and activating the antioxidant system[24]. It effectively extends the storage period of "Murcott "mandarins while maintaining acceptable quality and slowing down postharvest deterioration[25]. SA at various concentrations has significant effects in reducing fungal invasion compared with control during cold storage of "Kinnow" mandarins[26]. Using 0.3% SA on "Jinshayou "pummelo fruit can be a safe preservation method that maintains fruit quality, delays senescence, and extends the storage period for up to 90 days under room conditions[27]. The aim of this study is to utilize BPE as a safe edible coating a lone or incorporated with CHNPs and SANPs to preserve the quality of "Murcott "fruits and extend their storage period at 5ºC. This approach not only adds value to banana waste by using it in sustainable technologies but also reduces the reliance on synthetic fungicides for fruit preservation .

2. Materials and Methods

During the 2022 season, this study was conducted on "Murcott "tangerines (*Citrus reticulata* L) fruits taken from trees grown in a private orchard located in the El-Bostan region (30°46'10.0"N 30°19'12.9"E), El-Behera Governorate, Egypt. The tangerine trees were nine years old and budded on sour orang (*Citrus aurantium* L.) rootstock. The orchard was situated in sandy soil and utilized a drip irrigation system. The "Murcott "tangerine fruits were hand-harvested during the first week of March, when the hue angle was measured at 61.42 and the TSS/TA ratio was 13.56%)[28]. The fruits were carefully picked in the early morning from all sides of the trees, ensuring they were free from any physiological or pathological disorders. After harvesting, the fruits were washed using running tap water and then dipped in a solution of 1% (w/v) boric acid and Tween-20 for duration of 5 minutes. Finally, the tangerines were left to air-dry. Thus, the postharvest experiment had 9 treatments, as presented in table 1.

2.1.Preparation methods

The preparation of banana peel extract (BPE)

The preparation of BPE was carried out in accordance with the method outlined by [29]. Initially, the banana peel was dried in an oven at a temperature of 70˚C for a period of 3 days. Once dried, the peel was crushed into a powder form and blended thoroughly. Subsequently, 1000 mL of distilled water was mixed with 100 g (T2) and 200g (T3) the banana peel powder. The flask containing the mixture was then placed in a shaker machine (Heating, Model No KI-215), set at a temperature of 40 ˚C, and left to shake for duration of 48 hours. Following this, the mixture was filtered using Whatman filter paper No. 4.

2.1.1.Preparation of chitosan nanoparticles (CHNPs)

To prepare CHNPs, CH was dispersed in 1 % aqueous acetic acid. The pH was then adjusted to 5.5 using NaOH. Sodium tripolyphosphate solution (1 %) was added to the CH solution. The resultant suspension was kept at room temperature stirred for 40 minutes[20]. Examination of nanoparticle formation using transmission electron microscopy (TEM) was carried out.

2.1.2.Preparation of SA Nanoparticles

SA was purchased from Sigma-Adrich, and 10 mg of SA were dissolved in 10 mL 100% ethanol and solicited for an hour at ambient temperature (25 ◦C) using an ultrasonic power and frequency of 50 kHz (XUBA3Analogue Ul-ta-sonic Bath, Grant Company, Saint Joseph, MO, USA)

To prepare the coating solutions, Tween-20 (0.1%) was added to each solution as a surfactant. The fruits were then dipped in these solutions for a period of 5 minutes. Both treated and untreated fruits were allowed to air dry for 2 hours to ensure surface dryness. Each treatment consisted of 9 boxes of fruit, with 3 boxes allocated to each replicate. All boxes contained the same number of fruits (25 fruits per box). The boxes were stored at a temperature of (5±1°C) and a relative humidity of (90±1)%. Throughout the storage period, fruit quality parameters were assessed at intervals of 20 days.

Table 1. Postharvest treatments combinations of BPE, CHNPs, and SANPs

Postharvest treatments	Description
T1	Dipping in water (control)
T2	Dipping in BPE 10 % for 5 minutes
T3	Dipping in BPE 20 % for 5 minutes
T4	Dipping in CHNPs 2 % for 5 minutes
T5	Dipping in CHNPs 3% for 5 minutes
T6	Dipping in SANPs 0.25% for 5 minutes
T7	Dipping in SANPs 0. 5% for 5 minutes
T8	Dipping in BPE 10 %+ CHNPs 2 %+ SANPs 0.25% for 5 minutes.
T9	Dipping in BPE 20 %+ CHNPs 3 %+ SANPs 0. 5% for 5 minutes

2.2. Fruit quality assessments:

Physical characteristics:

Fruits decay % as the following equation:

Fruits decay % = {(A / B) × 100}

Where: A = Number of decayed fruits at time of sampling. B = Number of the initial fruits.

weight loss % as the following equation:

Weight loss % = [(A – B) / A] × 100.

Where: A = The initial weight. B = Weight at inspect date according to Elnagar et al [18].

Total loss = (weight loss) + fruits affected by green rot and brown rot. Rind color was determined by using a Minolta colorimeter type (CR-400/410) for the estimation of a, b and hue angle (h^o).,

Fruit firmness (kg/ cm^2) by using a digital pressure tester (pentromete), and juice weight % according to Elnagar et al. [18]

2.3. Chemical characteristics:

2.3.1. Total soluble solid (TSS)

TSS was determined in 10 ml, of filtered fruit juice by refractometer (Atago Co., Tokyo, Japan) as described by A.O.A.C. [30].

2.3.2. Titratable acidity (TA)

Fruit juice acidity for was estimated titrimetricaly by mixing the juice sample with 100 ml distilled water. The results were calculated as gm. per 100 ml of juice, [31]. Titratable acidity was measured as citric acid equivalent to g/100 ml, juice.

2.3.2. Ascorbic acid (vitamin C): The amounts of Ascorbic acid in juice samples were determine by the use of 2,6-dichlorophenol indophenol dye and 0.2% oxalic acid as a substrate and 5 ml. of filtered aliquot. It was calculated as mg/ 100 ml. of juice [32].

2.3.3. Respiration rate: Four fruits from each replicate were weighed, labeled, and assigned for respiration rate measurement using a closed-system approach. Each fruit was placed in an airtight glass flask of known volume. After packaging, the flask was tightly sealed and maintained at 5°C and 95% relative humidity (RH) for two hours. Both oxygen (O_2) and carbon dioxide (CO_2) concentrations inside the jar were monitored using a Servomex 1450C Food Package Analyzer (Crowborough, Sussex, UK). The respiration rate was calculated and expressed as ml CO_2/kg/h following the method described by [18] .

2.3.4. Enzyme activity

Superoxide Dismutase Activity (SOD): SOD activity was measured following a modified protocol from [33]. A 0.3 g sample of powdered material was suspended in 3 mL of child 0.05 mol/L phosphate buffer (pH 7.8). The sample mixture was centrifuged at 4°C and 12,000 rpm for 10 minutes, and the supernatant was collected as the enzyme solution (performed on ice). A reaction mixture was prepared by combining 2.95 mL of 130 mmol/L Met, 750 µmol/L NBT, 100 µmol/L EDTA-Na_2, 20 µmol/L riboflavin, and 0.05 mol/L pH 7.8 PBS in a test tube. Then, 50 µL of the enzyme solution was added to the reaction mixture. For the control group, 50 µL of PBS was substituted for the enzyme solution. The mixture was thoroughly shaken to ensure proper mixing. One tube was kept in darkness, while the other tubes were exposed to 4000 Lx light for 20 minutes. One unit of SOD is defined as the amount of enzyme that reduces the rate of nitro blue tetrazolium reduction by 50%.

POD activity was measured by the method of [34] the absorbance was recorded at 420 nm.

The determination of PPO activity was done according to [35] at 420 nm at 25°C.

Malondialdehyde (MDA) determination

A marker of fatty acid breakdown, thiobarbituric acid reacting substances (TBARS), was quantified in a precisely weighed 2.5 g of Murcott tangerine sample according to [36]. To establish a standard curve, solutions of a precursor compound (Sigma) were prepared with concentrations ranging from 0 to 2 mM (malondialdehyde equivalents), corresponding to 0 to 1 mM MDA. During the acidification and heating stage, the precursor compound was completely converted to MDA.

2.3.5. Total Phenolic content:

To extract phenolic compounds from mandarin, 10 g of fruits were homogenized with 60 mL of a solvent solution comprising 80% aqueous ethanol and 1% concentrated HCl. Following the method described by [37], the mixture was subjected to magnetic stirring for one hour under darkness at room temperature (25°C) and subsequently filtered. This process was repeated twice to ensure complete extraction. Excess ethanol was removed by heating the mixture at 37°C under vacuum in a rotary evaporator. The resulting aqueous extracts, referred to as crud extracts, were combined to obtain a final volume of known quantity. The TPC of the crude extract was determined spectrophotometrically at 760 nm using a UV-2401PC spectrophotometer (Shimadzu Scientific Instruments, Columbia, MD, USA). The Folin-Ciocalteu colorimetric method was employed to measure the TPC of mandarin fruits, following the procedure outlined by [38]. The TPC was expressed in mg/g using gallic acid as a standard.

2.4. Evaluation of sensory quality:

The Murcott tangerine fruit was subjected to sensory evaluation on the 3rd day after the storage period ended. To determine if there were any noticeable distinctions in terms of superficial and interior quality (such as appearance, firmness, ease of peeling, smell, and taste) between the treated Murcott tangerine and control fruits, the triangle discrimination test [39] was employed. A scale ranging from 1 to 10 was used for assessment.

Statistical analysis

The study was conducted using a factorial completely randomized design with three replications. The means of the treatments were compared using the least significant difference test (L.S.D.) at a significance level of 5%. All data were analyzed statistically following the methods described by [40] .

Results and Discussion

3.1. Physical characteristics of fruit

3.1.1. Decay percentage

The graph in Figure 1A shows how the percentage of decay in Murcott fruits increases over time during storage at 5°C for all treatments. It's important to note that all treatments significantly reduced decay compared to untreated fruits. Specifically, treatments T9 and T8 had the lowest decay percentages, while treatments T1 and T6 had the highest decay percentages throughout the storage period. The interaction between the postharvest treatments and storage periods had a notable impact on the decay of mandarin fruits. T9 and T8 consistently showed the lowest decay values compared to the control at various sampling times (20, 40, 60, 80, and 100 days. The decay of fruit during storage is mainly caused by weight loss, which leads to shriveling and deterioration. Coating treatments are known for their ability to reduce weight loss and effectively decrease the decay % of cactus pear fruits[41]. Dipping Murcott fruits in SA and CH 1% has been proven effective in reducing decay and controlling green mold in citrus [6,42]. Additionally, [29] found that spraying BPE on ripe banana fruits can extend their shelf life by approximately 2-3 days and reduce decay. Similarly, [4] pointed that citrus fruits are prone to rot and mycotoxin production, making them unsuitable for consumption. This susceptibility to pathogenic fungi attack is due to the high moisture, low pH, and nutrient-rich juice in citrus fruits.

3.1.2. Weight loss percentage

In Figure (1B), effect of different postharvest treatments on the weight loss percentage of Murcott fruits during cold storage at 5±1 ℃ is shown. As the storage period increased and different dipping treatments were applied, the weight loss percentage also increased. However, the fruits treated with T9 and T8 had significantly lower weight loss percentages compared to the other treatments. In fact, T9 exhibited the lowest weight loss percentage among all treatments, considering the interaction effect during different storage periods. Notably, the Control (T1), T6, and T4 treatments resulted in significantly higher weight loss percentages of the fruits. These findings support the observations made by [43], who found that uncoated lemons experienced maximum weight loss, while coated lemons maintained their weight well during storage. Additionally, [29]discovered that spraying ripe banana fruits with BPE reduced the weight loss percentage. Using SA and CH as postharvest treatments on Murcott fruits proved effective in reducing the weight loss (%) compared to untreated fruits[6,44]. This limiting in weight loss can be attributed to the coating acting as a semi-porous barrier that limit the motion of O_2, CO_2, moisture, and solutes. As a result, respiration rate, water loss, and oxidation reaction are decreased[45].

3.1.3. Total loss (%)

According to the data presented in Figure (1C), it is evident that the total loss percentage of Murcott Tangerine mandarin fruits generally increased with the progression of the storage period for all treatments. The main factors contributing to the deterioration of the fruits were increasing (weight loss), as well as green mold and brown rot. However, all coating treatments showed a significant reduction in the total loss percentage of Murcott fruits. Specifically, treatments T9 and T8 recorded the lowest total loss percentages compared to treatments T1 and T6 at different sampling times ranging from 10 to 100 days. These findings are consistent with the research conducted by[41,46], who reported an increase in decay percentage in Egyptian Banzahir lime and “Murcott” fruits during storage due to weight loss, resulting in shriveling and deterioration. The use of BPE (presumably BPE) and CHNPs exhibited similar effects to commercial wax, reducing the weight loss of the fruits. This reduction in weight loss can be attributed to the coatings acting as porous barriers opposed to O_2, Co_2, and moisture, thereby decreasing respiration, water loss, and oxidation reaction rates [47].

3.1.4. Rind color:

The findings from figure (1D) demonstrate that fruit color (h°) was affected by the dipping treatments and extended storage at 5±1 ℃. As the storage period progressed at 5 ℃, the h° values of the fruits generally decreased significantly, reaching their lowest point after 100 days of storage. However, all treatments were successful in reducing the decline of h° values in Murcott fruits. Treatments T9 and T8 had significantly higher h° values compared to the control or other treatments. This suggests a relationship between the postharvest treatments and storage periods. On the other hand, treatments T1 and T6 showed a significant increase in the lowest Hue angle during storage. These results are consistent with previous research on lime fruits, Orange, mandarin and 'Star Ruby' fruits Elnagar et al [18]. Additionally, it has been reported that BPE can effectively maintain color, flavor, and firmness during longer storage of bananas. The application of different edible coatings also helps slow down the change in Hue angle during storage compared to uncoated fruits. The lower in h° value is attributed to the ripening process, which involves a decrease in phenolic compounds content and the inhibition of enzyme activity responsible for carotene content.

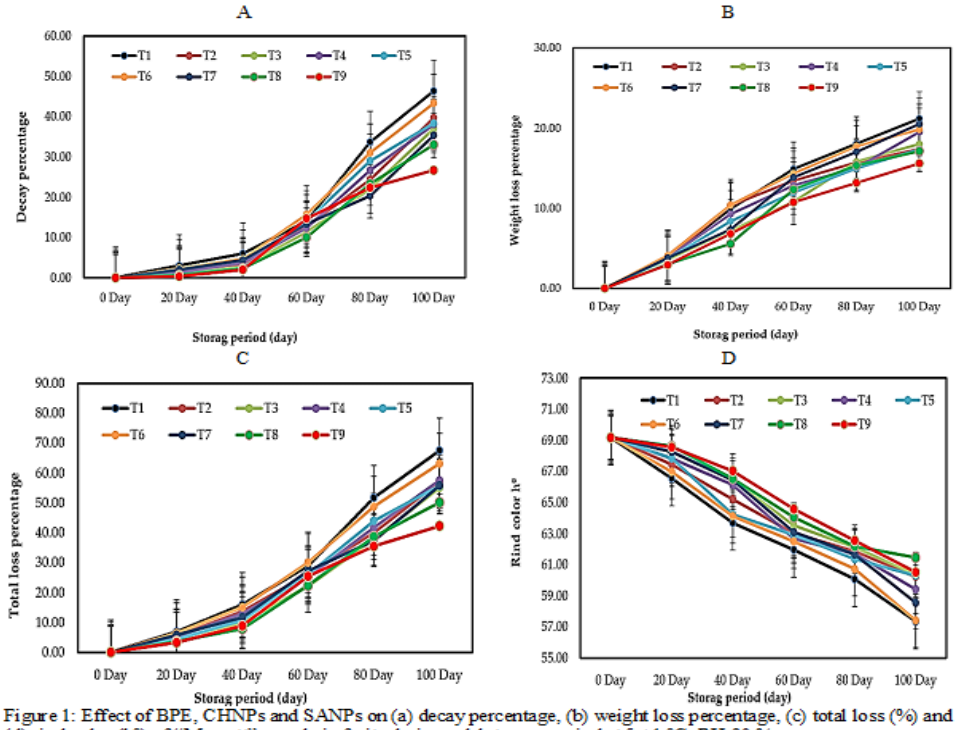


Figure 1: Effect of BPE, CHNPs and SANPs on (a) decay percentage, (b) weight loss percentage, (c) total loss (%) and (d) rind color (h°) of “Murcott” mandarin fruits during cold storage period at 5 ±1 °C, RH 90 %.

3.1.5. Fruit firmness (kg/cm^2)

Figure (2A) demonstrates that the firmness of Murcott fruits was influenced by different coating treatments and periods of cold storage at 5±1 °C. The data clearly show that as the storage period advanced, the fruit firmness exhibited a significant linear decline for all fruits stored at 5±1 °C. All treatments significantly increased the firmness of Murcott fruits, with treatments T9 and T8 resulting in the highest firmness values compared to other treatments, respectively. The interaction between storage periods and treatments revealed that dipping the mandarin fruits in T9 and T8 led to the highest firmness values at different sampling times during storage. Conversely, the lowest firmness was observed in T1, T6, T4, and T2 among all treatments. Similar findings were reported by [18], who found that CH coating significantly increased the firmness of Murcott mandarins after 90 days of cold storage at 5 °C. Additionally, [25] showed that dipping Murcott mandarin fruits in SA at 400 ppm effectively maintained fruit firmness compared to the control. [29] also demonstrated the significant effect of BPE on preserving acceptable color, flavor, and firmness during longer storage of bananas. The decrease in fruit firmness of Murcott mandarins can be attributed to excessive water loss, which leads to increased ethylene biosynthesis and a decrease in polygalacturonase activity during storage. Furthermore, the degradation of insoluble protopectin to more soluble pectic acid and pectin contributes to the decrease in firmness observed in many fruits[48,49]. The decline in firmness occurs as the pulp separates from the peel during the maturation process, particularly in mandarins[50].

3.1.6. Juice percentage:

As the concentrations of SA, CHNPs, and BPE increased, the rate of decrease in juice content for coated samples was lower. Specifically, treatments T9 and T8 exhibited a minor drop in juice content compared to all other treatments at the end of the storage period (Fig. 2B). On the other hand, treatments T1 and T6 showed a higher rate of juice decrease. These findings align with the results reported by [51] in their study on "Navel" orange fruits, where they found that the juice content of the fruits decreased with an increase in the storage period. CH coating significantly prevented the decrease in juice content of Murcott mandarins after 90 days of cold storage at 5 °C, as reported by [18]. Additionally, coating with oils and waxes has been found to be effective in preventing weight loss and juice loss, inhibiting microorganisms, slowing down aerobic respiration rate, and improving the appearance of fruits by providing gloss, according to studies by [52][37]. There is a relationship between weight loss percentage and juice percentage, where an increase in weight loss indicates a decrease in juice content during storage, as noted by [53].

3.2. Chemical characteristics of fruit

3.2.1. TSS percentage

TSS content serves as a crucial parameter for assessing the quality of mandarins and oranges. Fig. (2C) presents the impact of various dipping treatments on the TSS percentage of Murcott fruits during cold storage. Over the storage period, there were minor increments in the TSS percentage, reaching its peak after 100 days at 5±1 °C. All treatments effectively mitigated the rise in TSS percentage of Murcott tangerine fruits, with treatments T1, T6, T4, and T2 exhibiting the highest TSS values, respectively. It is important to note that non-climacteric fruits like mandarins undergo minimal changes in TSS, vitamin C, or acidity post-harvest, as mentioned by [54]. These findings are consistent with the results reported by [18] (2021), who observed slight increases in TSS during the 100-day storage of "Murcott "tangerine fruits in cold storage. Moreover, studies conducted by [27] on pummelo fruit treated with 0.3% SA demonstrated its significant ability to prevent changes in TSS content during storage periods. Similarly, [29] reported that bananas treated with BPE exhibited slower rates of TSS increase compared to the control. Conversely, treatments T9, T8, T3, and T5 recorded the lowest TSS values, respectively, in comparison to the control. Furthermore, these treatments demonstrated the lowest TSS content of Murcott tangerine fruits in relation to the interaction effect during different storage periods. The slight increase in TSS content during storage can be attributed to the breakdown of cell walls containing polysaccharides, particularly pectin and cellulose, due to the activity of cell wall degrading enzymes. This phenomenon has been previously reported by [43].

3.2.2. Total acidity percentage

Mandarins are known to contain significant amounts of organic acids, with citric acid being the main constituent of the juice. Figure (2D) displays the effect of different coating treatments on the total acidity percentage of "Murcott "tangerine fruits during cold storage at 5±1 °C. During storage at low temperatures of 5°C, the total acidity percentage of the fruits significantly decreased for all the different fruit treatments. The fruit coated with T9, T8, T2, and T3 showed the highest values of total acidity percentage after 100 days of storage. Conversely, the lowest values were recorded for T1 and T6 treatments during the storage period. When considering the interaction effect between the postharvest treatments and storage periods at 5±1 °C, it was observed that the highest total acidity percentage of "Murcott "tangerine fruits at different sampling times (20, 40, 60, 80, and up to 100 days) was associated with coating the mandarin fruits with T9 and T8 compared to the control. These findings are consistent with the results reported by [41] on Egyptian Banzahir lime, [55] on "Kinnow "mandarin, [56] on "Tango "mandarin, [57] on "Ortanique "mandarin, [27] on Pummelo fruit, and "[58] on "Kinnow "mandarin. These studies have shown that fruit acidity continuously decreases with increasing cold storage period at 5±2°C with 85-90% relative humidity for all treatments. However, the decrease in acidity was slightly less pronounced when the fruits were coated with substances such as Arabic gum, wax 10% K_2SiO_3, SA and CH compared to the control. The decreasing trend in fruit acidity with increasing storage period can be attributed to the oxidation of organic acids and their utilization in metabolic processes, as suggested by [59].

3.2.3. Ascorbic acid (mg/ 100 ml juice):

In Figure (3A), it is evident that the different treatments had an impact on the ascorbic acid content of Murcott tangerine fruits during cold storage at 5±1 °C. The overall trend showed a significant decrease in ascorbic acid values as the storage period progressed for all treatments, reaching the lowest values after 100 days of storage. However, all treatments effectively reduced the loss of ascorbic acid in the "Murcott "tangerine fruits during cold storage. While there were significant variations in ascorbic acid values among the treatments, fruits treated

with T9, T8, and T4 exhibited significantly higher ascorbic acid values compared to other treatments. This suggests that these treatments were successful in preserving ascorbic acid, possibly by reducing the respiration rate of the dipped fruits.

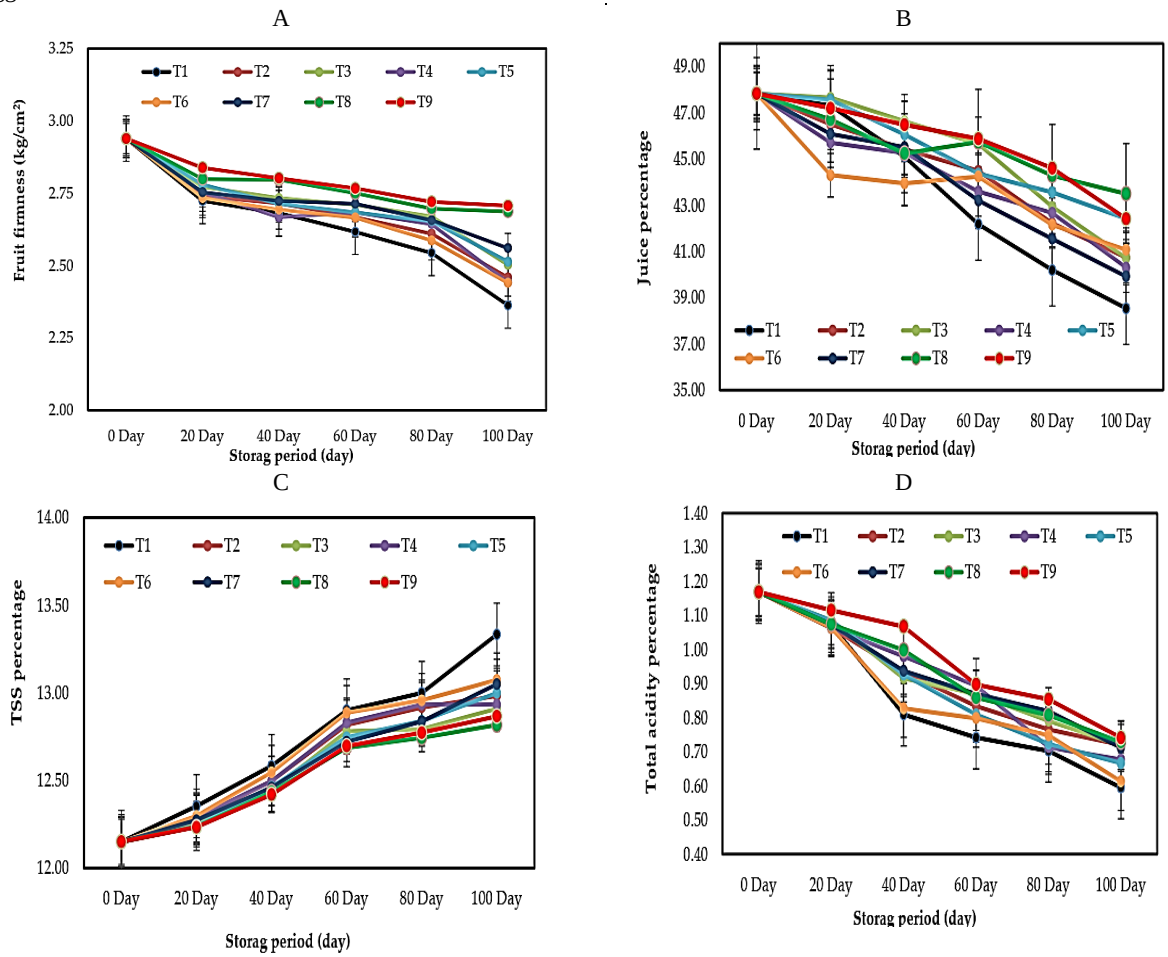


Figure 2: Effect of BPE, CHNPs and SANPs on (a) fruit firmness (kg/cm^2), (b) juice percentage, (c) TSS percentage and (d) total acidity percentage of Murcott mandarin fruits during cold storage period at 5 ±1 °C, RH 90 %.

Regarding the interaction between the different storage periods, T9 and T8 displayed the highest ascorbic acid content in Murcott tangerine fruits. Conversely, T1 treatment resulted in significantly lower ascorbic acid values. These results align with the findings of [43] and [41], who also observed a reduction in ascorbic acid levels during cold storage. Additionally, the use of edible coatings on tangerine fruits can partially limit gas exchange through the fruit peel and inhibit the action of ethylene during storage, thereby reducing the respiration rate. This inhibitory effect contributes to maintaining a better level of ascorbic acid, as reported by [60]. Similar findings have been reported in other studies. For instance, [26] found that "Kinnow" mandarin fruit treated with 4 mM SA exhibited the highest ascorbic acid, phenolic, and antioxidative activity contents during cold storage. [24] also observed an improvement in the ascorbic acid content of pummelo fruits after treatment with 1.5% CH compared to the control during storage at room temperature (20 ± 2 °C). In general, the decrease in ascorbic acid during storage can be attributed to the rapid conversion of L-ascorbic acid into dehydroascorbic acid in the presence of L-ascorbic acid oxidase, as suggested by [60].

3.2.4. Respiration rate (ml CO_2. Kg^{-1} h^{-1})

All treatments had a significant effect on reducing the respiration rate of Murcott tangerine fruits stored at 5±1 °C (Fig. 3B). Initially, the respiration rate was measured at 6.24 ml CO_2 kg^{-1} h^{-1}, and it decreased during the first 20 days of storage at 5±1 °C. However, after this initial period, the respiration rate gradually increased with the progress of the storage period, regardless of the treatments used. It should be noted that "Murcott " tangerines, like other mandarin fruits, are non-climacteric, meaning their respiration rate does not increase with ripening and senescence. Among the treatments, T9, T8, T3, and T7 resulted in significantly lower respiration rates during storage compared to the other treatments. Additionally, fruits treated with T9, T8, T3, and T7 had the lowest respiration rate values at different sampling times (20, 40, 60, 80, and up to 100 days of storage) compared to the control group. These findings align with a study by [61], which demonstrated that coatings significantly reduce the respiration rate of mandarin fruits during cold storage. Edible coatings restrict gas exchange through the fruit peel, inhibit the actions of ethylene, and reduce the respiration rate, thus delaying the aging

process of the fruits. This is supported by studies conducted by [62], [63], [59], [64], [65].

In this study, the treatments applied successfully decreased the respiration rate of "Murcott "tangerine fruits during cold storage. This reduction in respiration rate has the potential to preserve fruit quality and extend their shelf life. Throughout the storage period at 5±1 °C, all treatments resulted in a significant decrease in the respiration rate of "Murcott "fruits, which was measured in ml CO_2 kg^{-1} h^{-1}. The initial respiration rate of the fruits was measured at 6.24 ml CO_2 kg^{-1} h^{-1}, and during the first 20 days of storage at 5±1 °C, the respiration rate of Murcott tangerines decreased. However, as the storage period progressed, the respiration rate gradually increased regardless of the treatments, as depicted in figure 10. It is worth mentioning that "Murcott "tangerines belong to the non-climacteric group of mandarin fruits. Unlike climacteric fruits, these tangerines do not undergo a noticeable increase in respiration rate during ripening and senescence, as observed in a study by [66]. Among the treatments used in this study, namely T9, T8, T3, and T7, significantly lower respiration rates were observed during the storage period compared to the other treatments. These four treatments also recorded the lowest respiration rate values at different sampling times (20, 40, 60, 80, and up to 100 days of storage) compared to the control group. These findings align with the study conducted by [61], which demonstrated that the application of coatings significantly reduced the respiration rate of mandarin fruit during cold storage. Another study by [24,67] found that treating pummelo fruit with 1.5% CH significantly slowed down the respiration rate. This outcome was linked to the increased presence of active antioxidant enzymes, such as SOD, catalase (CAT), ascorbate peroxidase (APX), and glutathione reductase (GR). Additionally, according to [43], coating mandarin fruits with either CH or coconut oil led to a reduction in the initial respiration rate by nearly half or even less. The application of an edible coating partially limits gas exchange through the fruit peel, inhibits the effects of ethylene, and decreases the respiration rate, thereby slowing down the aging process of the fruit. This aligns with the findings of previous studies by [68]. In conclusion, the treatments applied in this study effectively reduced the respiration rate of Murcott tangerine fruits during cold storage, potentially extending their shelf life and preserving fruit quality.

3.2.5. MDA content, SOD, POD, and PPO Activities

The present findings indicate that the MDA content of "Murcott "tangerine fruits varied significantly among different treatments at the end of the storage period (Fig. 4A). Notably, treatments T9, T8, and T3 exhibited positive effects in reducing MDA levels, suggesting improved fruit storability and quality.

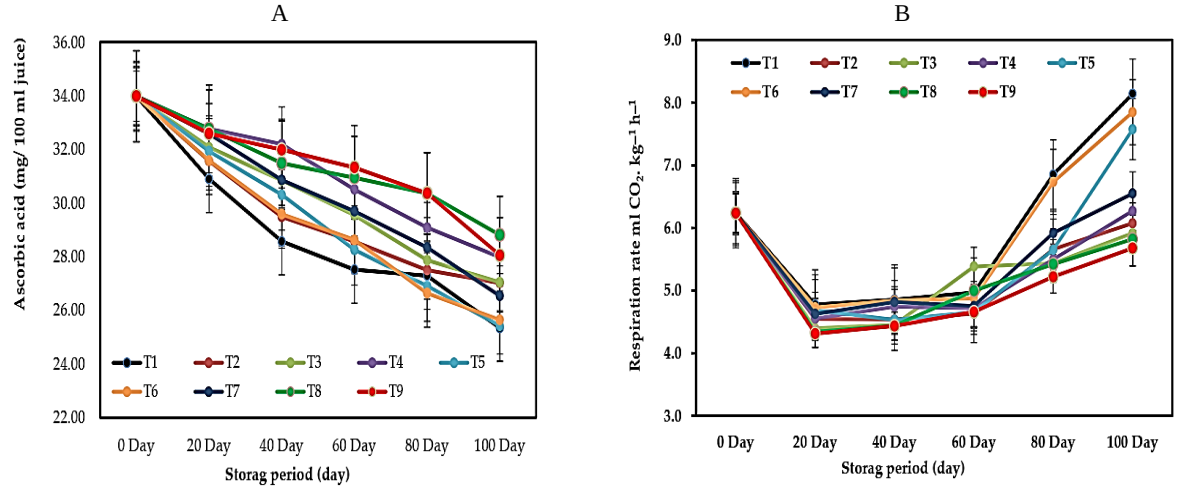
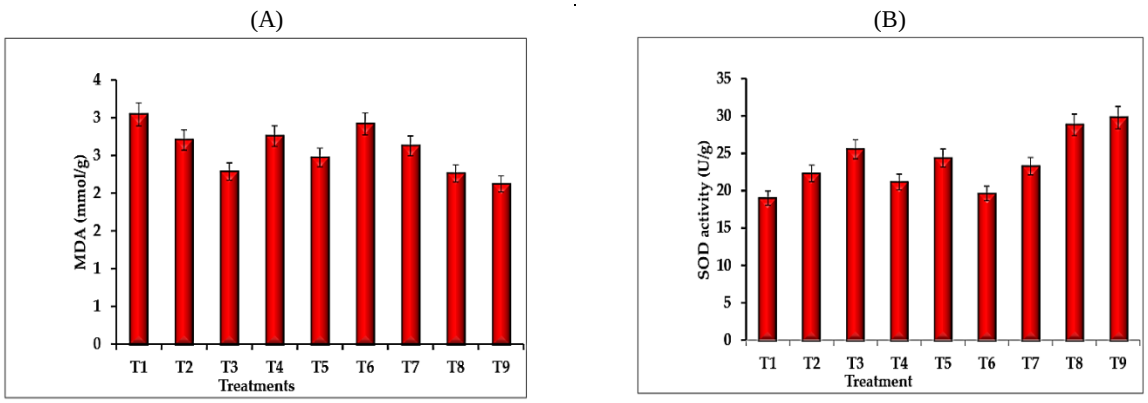


Figure 3: Effect of BPE, CHNPs and SANPs on (a) Ascorbic acid (mg/ 100 ml juice) and (b) respiration rate (ml CO_2. kg^{-1} h^{-1}) of Murcott mandarin fruits during cold storage period at 5 ±1 °C, RH 90 %.



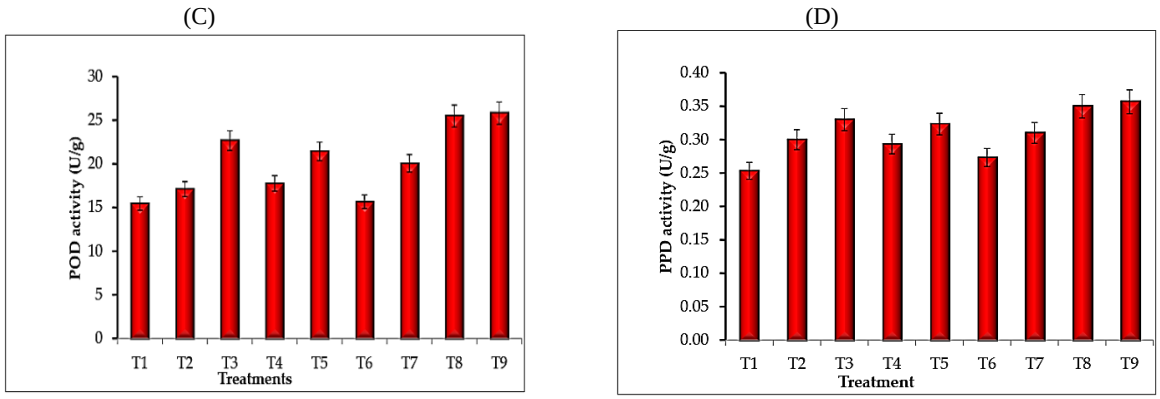


Figure 4 (A- D): Effect of BPE, CHNPs and SANPs on MDA content (A), SOD (B), POD (C), and PPO (D) activities of Murcott mandarin fruits during cold storage period at 5 ±1 °C, RH 90 %.

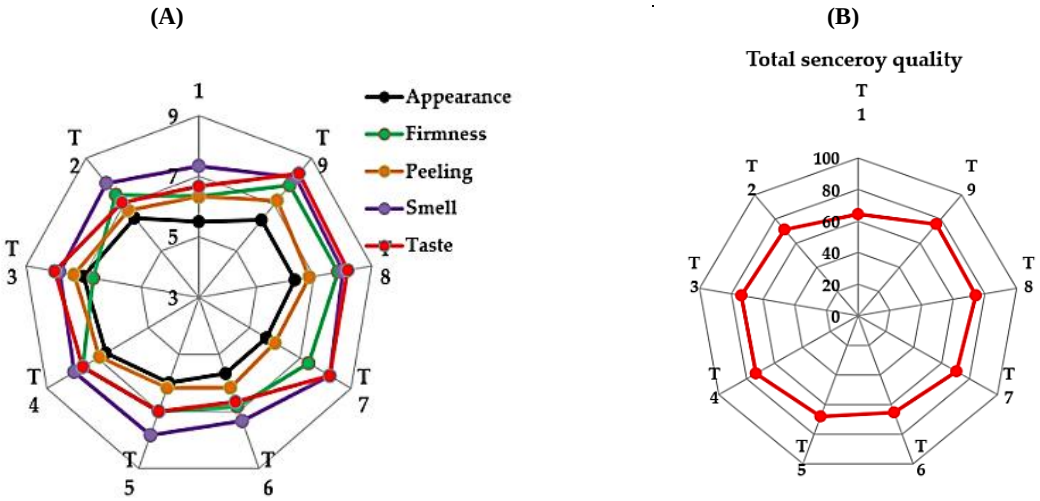


Figure 5: Effect of BPE, CHNPs and SANPs on Sensory quality of Murcott mandarin fruits during cold storage period at 5 ±1 °C, RH 90 %.

Conversely, treatments T1, T6, and T4 recorded the lowest MDA content at the end of the storage period. MDA is commonly used to measure oxidative cell damage during storage and assess fruit ripening progress, with levels increasing as ripening progresses [69]. Furthermore, the treatments involving banana waste, CH and SANPS had a notable impact on the activities of SOD, POD, and PPO enzymes. All the treated fruits exhibited higher levels of SOD, POD, and PPO activities compared to the control group (Fig. 4 B, C and D). Among the treatments, T9, T8, and T3 demonstrated the highest enzyme activity. Conversely, T1 and T6 displayed the lowest SOD, POD, and PPO activities compared to the other treatments at the end of the shelf life. Similar findings were reported by [70] and [71], who observed that the application of an edible coating increased CAT activity and decreased PPO activity, consequently prolonging the shelf life and preserving fruit quality during storage. It is plausible that there exists a connection between the total antioxidant content of treated fruits and various quality characteristics such as weight loss, decay percentage, respiration rate, TSS, and PPO activity. The combination of coating treatments may mitigate the effects of enzymes such as PPO by obstructing the fruit's ability to respire, thereby increasing phenol content [72,73].

3.3. Sensory quality

Figure (5 A and B) illustrates a significant reduction in sensory quality attributes (Appearance, Firmness, Peeling, Smell, and Taste) of "Murcott "tangerine fruits after 100 days of storage at a temperature of 5°±1° C, with an additional 6 days as shelf life. However, all treatments had a positive impact on improving these sensory qualities. Specifically, treatments T4 and T3 exhibited the highest improvement in appearance, while T9 and T8 showed the greatest enhancement in firmness. T3 and T9 resulted in easier peeling, and the best taste and smell were observed in T9, T8, and T7. Moreover, after 6 days as shelf life, T9 and T8 demonstrated better retention and acceptance of sensory attributes (appearance, peeling, aroma, and taste) compared to uncoated fruits and other treatments. The highest score for total sensory quality was observed in T9 and T8. These findings align with previous studies by El- [74] and [75], which reported that coated Pomelo exhibited significantly higher glossiness, firmness, and uniformity. Additionally, Jojoba oil and CH treatments scored the highest in sensory quality for "Murcott "tangerine fruits after the storage period at 5°±1° C and room conditions, with an additional 3 days as shelf life, compared to the control [18]. [26] also found that the application of 4 mM SA effectively preserved the quality of "Kinnow" mandarin fruit during storage, making it suitable for long-term cold storage. The sensory quality of citrus fruits can be improved during storage by

applying surface coatings, which help in reducing water loss, enhancing the external skin layer, and regulating the permeability of O_2 and CO_2 gases, thereby reducing transpiration rates [76]; [77], and [78].

5. Conclusions

The immersion of Murcott fruits in a composite film BPE (20%), CHNPs (3%), and SANPs (0.5%) for 5 minutes resulted to improving fruit quality. This treatment led to a decrease in decay percentage, weight loss percentage, and total loss percentage, while maintaining fruit firmness, color (h^o), total acidity, and ascorbic acid levels. Additionally, it effectively reduced the respiration rate. Furthermore, the sensory quality of Murcott fruits was well-preserved. On the other hand, the treatments with banana waste, CH and SANPs exhibited higher levels of SOD, POD, and PPO activities. In conclusion, our findings demonstrate that the utilization of (BPE 20% + CHNPs 3% + SANPs 0.5%) enhances the quality and storage capability of Murcott fruits when subjected to cold storage.

6. Conflicts of interest

There are no conflicts to declare.

7. Formatting of funding sources

This work has not received any funding.

Acknowledgments: The authors extend their appreciation to all members of the faculty of agriculture in Cairo, Al-Azhar University for providing support for this study.

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