Preharvest melatonin and postharvest xanthan gum coating maintain the quality of Orlando tangelos during storage

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Abstract

This study aimed to determine the effects of preharvest spraying of melatonin and postharvest immersion in xanthan gum on the quality and postharvest performance of Orlando tangelo mandarin fruits. After selecting suitable and uniform trees, melatonin foliar spraying was performed at three different concentrations: 0,100 µM, and 200 µM. Foliar spraying was performed one month before harvest and was repeated three times at weekly intervals. Furthermore, the fruits were immersed in two different concentrations of xanthan gum (0.1% and 0.2%) postharvest, these fruits were stored in a cold room at of 5 ± 1 °C. Evaluation of fruit characteristics was carried out at the time of harvest and after 45 and 90 days of cold storage. The results showed that foliar spraying of melatonin at a concentration of 100 µM showed the highest weight and pulp of the fruit. Furthermore, melatonin treatment resulted in higher levels of ascorbic acid and increased fruit acidity compared to the control. During storage, fruits treated with melatonin and xanthan coatings showed better quality than those of the control. At the end of the experiment, the lowest weight loss was observed in fruits treated with 200 µM melatonin + 0.1% xanthan. The highest ascorbic acid content was observed in the 100 µM melatonin +0.1% xanthan. The maximum antioxidant activity was observed in 100 µM and 200 µM +0.1% xanthan and also 100 µM melatonin alone.

In general, the findings suggest that preharvest foliar spraying and the postharvest application of xanthan coatings can be effective strategies for maintaining Orlando tangelo quality during cold storage.

Keywords: xanthan gum, spraying, melatonin, citrus, storage

Introduction:

Citrus is one of the most important fruit trees in several countries, including Iran. Citrus fruits contain abundant beneficial phytochemicals, including vitamins A, C, and E, mineral elements, flavonoids, coumarins, limonoids, carotenoids, pectins, and other compounds. These compounds contribute to the high nutritional value in humans (Saini et al. 2022). While there are various tangerine species, tangelo mandarin (*Citrus×tangelo*) has become a significant player in the citrus industry (Traore et al., 2023). Orlando tangerine is a hybrid of the Duncan grapefruit and Dancy tangerines. It is characterized by large, round, medium-ripe fruits that are juicy and fleshy, and have a relatively high seed count. In addition, they exhibited good heat resistance. It is estimated that 20-30% of fresh fruits are lost after harvest. This is mainly because of its perishable nature. To mitigate this, cold storage is used to delay ripening changes including ethylene formation, softening, pigment changes, respiration rate, acidity changes, and weight loss. However, because citrus fruits have a subtropical nature, they are susceptible to chilling when stored at low temperatures. Consequently, appropriate postharvest treatments in conjunction with cold storage are necessary to ensure the preservation of fruits at the desired levels of quality.

Melatonin, a natural endogenous plant hormone found in various crops, has been increasingly recognized for its positive effects on postharvest fruit preservation. Studies have shown that melatonin treatment can improve the content of bioactive compounds and the antioxidant activity in different fruits. Moreover, the environmentally friendly use of natural substances such as melatonin has emerged as an important approach to modulate the biosynthetic pathways that influence fruit quality during the ripening and postharvest stages (Arabia et al. 2022). In pomegranate trees of the "Mollar de Elche" cultivar, melatonin treatment has shown promising results in increasing fruit quality traits, such as anthocyanin and phenolic content, as well as overall fruit quality during storage. Treatment with preharvest melatonin at concentrations of 0.1 or 1 mM has demonstrated a significant impact on yield and quality improvement in pomegranate fruits during both harvest and postharvest storage (Medina-Santamarin et al. 2021). These findings highlight the potential of melatonin as a beneficial treatment for improving fruit quality and preservation, providing opportunities to optimize postharvest strategies and maintain fruit freshness and nutritional value. (Arabia et al. 2022; Lorente-Mento 2021).

Xanthan gum (XG), produced by *Xanthomonas campestris*, is generally recognized as safe (GRAS) by the Food and Drug Administration (FAO 2020). Natural hydrocolloid-based edible coatings and films offer additional protection to fresh or blanched fruits and vegetables. Edible coatings made from natural gums show promise for improving the quality and extending the shelf life of fruits and vegetables (Salehi, 2020). Edible coatings, particularly those enhanced with xanthan gum, play a crucial role in preventing quality deterioration by selectively regulating gas exchange between food and its external environment. This application not only extends the shelf life but also preserves the overall appearance and quality of fruits during storage (Tripathi et al., 2021). In a study on guava fruit, a mixture of 1% xanthan gum and 0.2% chitosan nanoparticles, used as a coating, was reported to improve the overall quality of guava fruits during long-term cold storage and extend their shelf life (Gad and Zagzog, 2017). Recently, Rastegar et al. 2024 reported that the postharvest application of melatonin and melatonin combined with γ-aminobutyric acid (GABA) plays an effective role in mitigating chilling damage in Orlando mandarin fruits stored at 3 ± 0.5 °C for 90 days. However, the literature lacks reports on the effects of preharvest application of melatonin and postharvest immersion in xanthan gum on the quantitative and qualitative characteristics of Orlando mandarin fruit. In light of the importance of preserving citrus fruits and the potential benefits of melatonin and xanthan gum-based edible coatings, this study aims to investigate the combined effect of preharvest melatonin treatment and postharvest xanthan gum-based edible coatings on the postharvest quality and preservation of Orlando tangerine fruits during cold storage. By examining the physicochemical and biochemical attributes of the treated fruits, we aim to provide valuable insights into the novel strategies for enhancing the postharvest quality and extending the shelf life of Orlando tangerine fruits, thus contributing to the citrus industry's efforts to minimize postharvest losses and meet consumer demands for high-quality fruits.

Materials and Methods

Fruit spray and harvest

In a citrus orchard located in Rudan city, an experiment involving foliar spraying of melatonin was conducted on 9-year-old Orlando tangerine trees (*Citrus paradisi* × *C. reticulata*). The experiment consisted of three melatonin concentrations: 0 (distilled water as a control treatment), 100 µM, and 200 µM. The spray was applied one month before harvest and repeated three times with a one-week interval between each application. Each treatment included 3 replicates and each replicate included one tree. Upon reaching commercial maturity in December, the fruits were harvested and transported to the laboratory for evaluation of their quantitative and qualitative characteristics. The collected fruits were partitioned into two distinct groups for the purposes of analysis. The initial group was subjected to an assessment in order to examine the impact of pre-harvest melatonin application on the quality attributes of the fruit. The subsequent group was utilized to investigate the post-harvest treatment involving xanthan, as well as the subsequent storage conditions for the fruits.

Preparation of xanthan gum solution

To prepare the xanthan gum solution, the gum was gradually dissolved in water and stirred for 30 min at room temperature. The resulting solution was then refrigerated for 24 h. Two concentrations of xanthan gum were prepared: 0.1% and 0.2%.

Covering fruit and storage

Fruit of the second category was subjected to disinfection with 0.05% sodium hypochlorite for one min, followed by washing with distilled water. The fruits were coated with xanthan gum solution using the immersion method. The fruits were immersed in the solution at room temperature for 5 min. The treatments and their abbreviations used in the experiment are shown in Table 1. After the surface coating was completely dried, the coated fruits were transferred to a fruit basket and stored for 90 days at 5 ± 1 °C. The desired factors and characteristics were evaluated after 45 and 90 days of storage to assess any changes or effects resulting from the treatments (Fig 1).

Table 1. Treatments and their abbreviations used in the experiment

Treatments	Abbreviation
Distilled water	Control
Melatonin 100 µM	M 100 µM
Melatonin 200 µM	M 200 µM
Xanthan gum 0.1%	XG 0.1%
Xanthan gum 0.2%	XG 0.2%
Melatonin 100 Mm+ Xanthan gum 0.1%	M 100 Mm+ XG 0.1%
Melatonin 200 µM + Xanthan gum 0.1%	M 200 µM + XG 0.1%
Melatonin 100 µM + Xanthan gum 0.2%	M 100 µM + XG 0.2%
Melatonin 200 µM + Xanthan gum 0.2%	M 200 µM + XG 0.2%

Fig 1. Different stages of experiment and measurement of fruit traits.

Length and diameter of the fruit

In order to quantify the dimensions of the fruit, a sample of 10 fruits was randomly selected from each treatment. The length and diameter of the selected fruits were then measured using a metal ruler, and the recorded measurements were expressed in centimeters.

Weight of the fruits

The weight of the fruit was assessed using a digital scale (SHS, Japan) with a precision of 0.1 grams. At the time of harvest (0 days), the weight of the fruit was assessed using a digital scale with a precision of 0.1 g.

Fruit volume

A graduated cylinder was used to measure fruit volume. A certain portion of the graduated cylinder was filled with water and the fruit was placed in it. The change in the water level indicated the volume of the fruit (Omid et al., 2010).

Weight loss

Weight loss was determined by comparing the weight of the fruit at the time of harvest (0 days) with the weight measured on sampling days 45 and 90 during the storage period. The weight of the fruits was recorded using a digital scale with an accuracy of 0.1 gr. The percentage weight loss was calculated using the following formula:

Percentage of weight loss = [(Initial weight - Final weight) / Initial weight] x 100

The measurements and calculations were conducted according to the methodology described by Dong and Wang (2018).

Total Soluble Solids (TSS)

To measure the total soluble solids (TSS) in fruit juice, a few drops of tangerine juice were placed on the screen of a digital refractometer (DBR95, Taiwan). The refractometer measured the TSS in Brix degrees.

Titratable Acidity (TA)

For TA quantitation, 5 mL of fruit juice was mixed with 20 mL of distilled water and titrated with 0.1 N NaOH to pH 8.2. The result was expressed as a percentage of citric acid.

Ascorbic acid content

A titration method was employed to measure ascorbic acid content. A small volume (0.05 ml) of fruit juice was mixed with 5 ml of 1% cold metaphosphoric acid. Subsequently, a solution containing indophenol (sodium-2,6-dichlorophenol-indophenol) was titrated until a purple color appeared. The amount of titrant used was indicative of ascorbic acid content, which was expressed as milligrams per 100 ml of juice (Bor et al., 2006).

Total Phenol

The total phenol content of the fruit juice was measured using the Folin-Ciocalteu reagent. A methanolic extract was prepared by mixing tangerine juice (0.5 ml of tangerine juice with 3 ml of 85% methanol and refrigerating for 24 h. Next, 60 μL of the methanolic extract was mixed with 300 μL of Folin's reagent (1:10), followed by the addition of 240 μL of 7% sodium carbonate. Following a two-hour period in darkness, the absorbance of the samples was measured using a microplate reader (Biotek model EPOCH2) at a specific wavelength of 760 nm (Ordonez et al.,2006).

Total flavonoid

Total flavonoid content was determined by adding 180 μL 85% methanol, 12 μL 10% aluminum chloride, 12 μL potassium acetate, and 336 μL distilled water to 60 μL methanolic extract. The mixture was left at room temperature for half an hour and the absorbance was measured with microplate reader at a wavelength of 415 nm.

Determination of the Antioxidant Activity

The scavenging activity of the DPPH (2, 2-Diphenyl-1picrylidrazil) radical was assessed using the method outlined by Brand-Williams et al. (1995) with some changes. To carry out the test, 30 μL of the methanolic extract was mixed with 150 μL DPPH solution, which contained 0.025 g DPPH in 100 mL of 85% methanol. The mixture was then left in the dark at room temperature for 40 min, and a microplate reader was used to measure the absorbance at 517 nm.

Statistical Analysis

The experiment was conducted as a factorial design in a completely randomized block design. Statistical analyses were performed using the SAS version 9.4. Average data were compared using the LSD test at a significance level of 5%. Graphs were plotted using Excel 2019 software.

Results and Discussion

Effect of melatonin pre-harvest spray on fruit characteristics at harvest time

The application of melatonin at a concentration of 100 μM resulted in a significant increase in the weight and pulp of the fruit compared to the control group. No significant effects were observed on other physical characteristics of the fruit. Ascorbic acid at 200 μM and titratable acid at both concentrations showed a significant increase compared with the control, but other traits were not affected by melatonin did not show any significant effects on fruit size and volume. This implies that melatonin application did not have a noticeable impact on the overall size and volume of Orlando tangelo (Table 2). However, it should be noted that the effect of melatonin on fruit characteristics can vary depending on the fruit variety and growing conditions. Melatonin plays a role in various physiological processes related to fruit enlargement and crop yield. Liu et al. (2019) showed that melatonin contributes to the enlargement of pear fruits. Similarly, in pomegranate trees treated with 0.1 mM melatonin, Medina-Santamarin et al. (2021) observed an increase in crop yield, including the number of fruits per tree and overall weight per tree. This effect was attributed to the improved development of the aril part of the fruit. The effects of melatonin on apple growth and ripening were investigated by Verde et al. (2022). This study demonstrated that melatonin treatment results in an increase in apple fruit size and weight. Additionally, in a separate study involving blackberries, the external application of melatonin increased the number, size, weight, and color of fruits (Verde et al. 2022). The observed effects of melatonin on fruit size and weight can be attributed to its ability to promote cell growth and enlargement, as has been reported by Zhao et al. (2023). Collectively, these studies suggest that melatonin has the potential to positively influence fruit characteristics, such as size, weight, and yield of various fruits. However, it is important to note that the specific effects of melatonin can vary depending on fruit species, cultivar, and experimental conditions.

Our findings revealed that the application of a high concentration of melatonin led to a significant increase in the ascorbic acid content of the fruit compared with both the control and a lower concentration of melatonin. This indicates that higher concentrations of melatonin, when applied by foliar spraying, have a pronounced effect on increasing the ascorbic acid levels in Orlando Tangelo mandarin fruits. Ascorbic acid, also known as vitamin C, is a vital antioxidant that plays a crucial role in various physiological processes and contributes to the nutritional quality of fruit. In addition to the increased ascorbic acid content, the treated fruits also exhibited a higher titratable acidity than the control. This suggests that melatonin application, particularly at higher concentrations, influenced the acidity of the fruit. The higher titratable acidity observed in melatonin- treated fruits indicates the possible impact of melatonin on the acid balance of the fruit. Fruits treated with melatonin 200 200 μM showed higher TSS than control. Similarly, it has been shown that in a study focusing on pear fruit, the foliar application of melatonin at a concentration of 100 μM resulted in an increase in the total soluble solids (TSS) of the fruit (Table .3)

Table 2. The effect of melatonin foliar spraying on the physical characteristics of Orlando tangelo fruit

Treatments	Fruit weight (g)	Peel weight (g)	Pulp weight (g)	Water volume (ml)	Fruit length (cm)	Fruit diameter (cm)	Fruit volume
Control	168 ^{b}	35.5 ^{a}	130 ^{b}	83.3 ^{a}	6.3 ^{a}	6.76 ^{a}	147 ^{a}
Melatonin 100 μM	218 ^{a}	48 ^{a}	166 ^{a}	90 ^{a}	6.4 ^{a}	6.7 ^{a}	182 ^{a}
Melatonin 200 μM	163 ^{b}	37.5 ^{a}	126 ^{b}	86 ^{a}	6.1 ^{a}	6.6 ^{a}	179 ^{a}

In each column, the numbers with the same letters are statistically not significantly different from each other at the 5% probability level.

Table 3. The effect of melatonin foliar spraying on the biochemicals characteristics of Orlando tangelo fruit

Treatment	Antioxidants (%)	Flavonoid (mg/gFW)	Phenol (mg/gFW)	Ascorbic acid (mg/100 ml)	pH	TA (%)	TSS (%)

Control	79[bc]	3.88[a]	9[ab]	56[de]	3.32[c-d]	0.39[c]	9.5[c]
Melatonin 100 μM	82.1[ab]	3.56[a]	9.12[a]	62[d]	3.43[a-c]	0.52[a]	9.7[c]
Melatonin 200 μM	76[c]	3.69[a]	7.63[b]	89[a]	3.18[f]	0.51[a]	10.4[bc]

In each column, the numbers with the same letters are statistically not significantly different from each other at the 5% probability level.

Postharvest study

Weight loss

After 90 days of storage, in the sample treated with 200 μM melatonin + 0.1% xanthan, as well as the sample treated with 0.2% xanthan alone, minimal weight loss was observed. This decrease in weight loss was found to be statistically significant when compared to the control group (Fig. 2).

Weight loss in fruits can occur because of water loss caused by changes in surface transfer resistance against water vapor, transpiration, and the rate of fruit respiration, as described by Shahid and Abbasi (2011). To prevent weight loss, edible coatings are found to be effective in minimizing transpiration. Edible coatings create a barrier on the surface of the fruit, partially or completely covering the openings and small pores, thereby reducing the gas exchange and transpiration (Vignesh and Nair, 2019). The ability of some coatings to limit water loss and maintain fruit weight during storage has been well documented (Kittur et al., 2001). The use of guar gum coating in tomatoes controlled the fruit weight loss, which is similar to the results obtained in our study (Ruelas-Chacon et al., 2017). The epidermal cell and cuticle layers play a role in reducing water loss (Khunk et al., 2020). Therefore, the lower weight loss observed in fruits treated with melatonin than in the control group can be attributed to the effect of melatonin in increasing the thickness of the cuticle (Bal, 2019). Pre-harvest melatonin treatment during cherry fruit growth in trees has also been shown to reduce fruit weight loss during cold storage (Carrión-Antol, 2022). In another study, postharvest application of melatonin delayed weight loss and fruit rot in blueberries (Shang et al., 2021). Additionally, treating pear slices with xanthan gum and calcium ascorbate was found to reduce fruit weight loss compared to the control group, a finding that aligns with our results (Guccione et al., 2023).

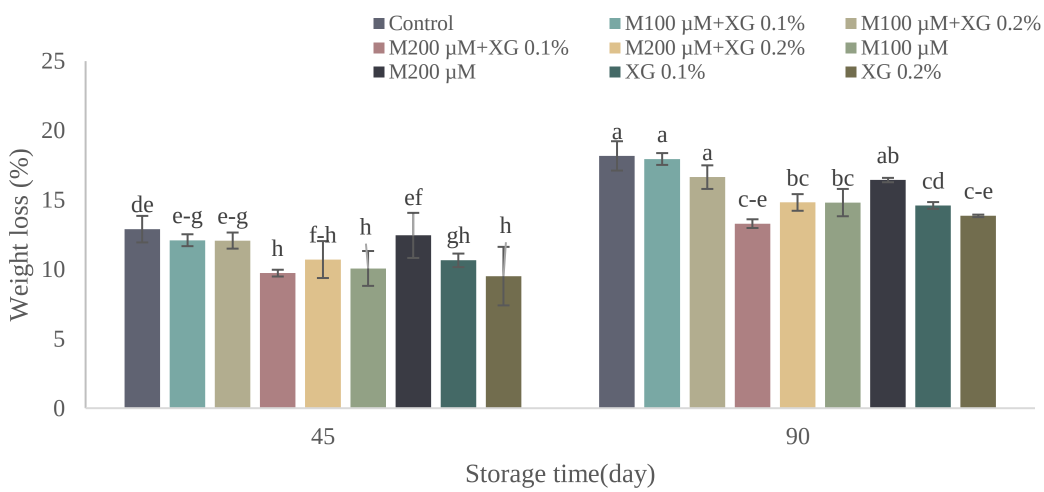
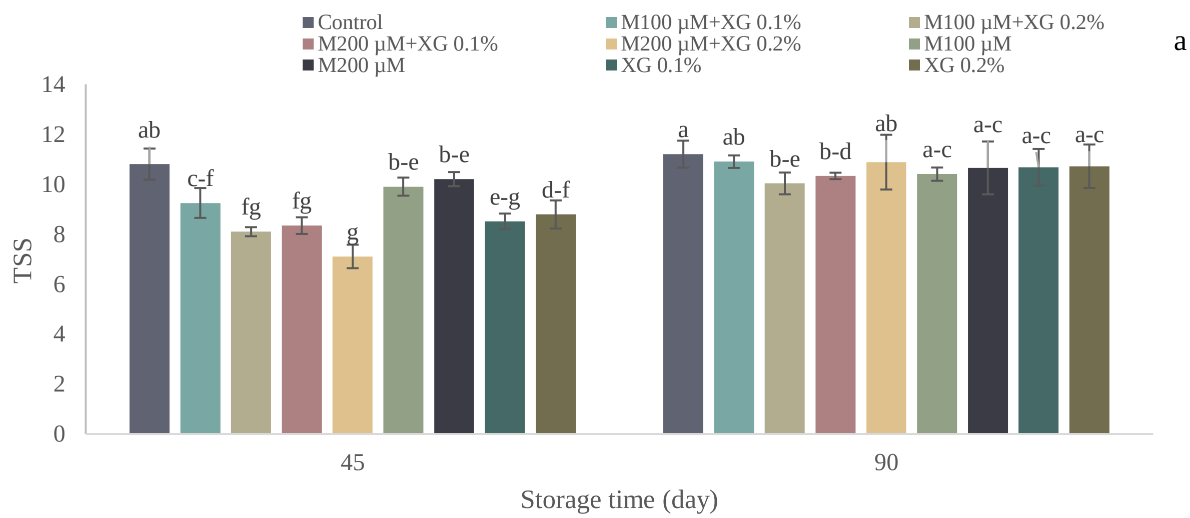


Fig. 2- The effect of different levels of melatonin spray and xanthan gum immersion on the weight loss percentage of Orlando tangelo

Total soluble solids (TSS) and titratable acidity (TA)

During fruit storage, the lowest TSS was observed in melatonin 100 µM+ xanthan 0.2% and melatonin 200 µM+ xanthan 0.1% treatments, which was significantly lower than that in the control and other treatments (Fig. 3 a). In a study focusing on pear fruit, the spray of melatonin at a concentration of 100 µM). Furthermore, composite coatings prepared from xanthan gum and lemongrass essential oil were found to be effective in maintaining the soluble solid content (SSC) and reducing oxidative stress in Kinnow mandarin fruits during long-term storage, as reported by Bajaj et al. (2024). This indicates that xanthan gum and lemongrass essential oil coatings can preserve the nutritional quality of fruit, which includes TSS parameters. In grapes, melatonin treatment increased the content of soluble solids, as demonstrated by Xull et al. (2017). In contrast, the combination of xanthan gum and olive oil in grapefruits reduced the accumulation of total soluble solids and total sugars, as reported by Baraiya et al. (2016). These findings highlight the potential role of different treatments, including melatonin and coatings, in modulating sugar content and TSS parameters of fruits.

The results showed that melatonin 100 µM+ xanthan 0.2%, melatonin 200 µM+ xanthan 0.2% and xanthan 0.1% treatments exhibited a significantly higher titratable acid (TA) content (Fig. 3b). In tomato fruits, the combined effect of coatings based on whey protein isolate, xanthan gum, and clove oil resulted in less degradation of titratable acidity compared to uncoated samples, as reported by Kumar and Saini (2021). This indicates that the coatings helped maintain the titratable acidity of the tomatoes. Furthermore, in a study involving preharvest treatments in strawberries, including melatonin application, Xia et al. (2020) found that treatments applied one week before harvest resulted in fruit with higher total soluble solids (TSS) and lower titratable acidity (TA). This suggests that the timing of treatment can influence the acid content of the fruits. It is important to consider the specific fruit species, treatment conditions, and experimental context when interpreting the effects of melatonin and other treatments on titratable acidity. More research is needed to fully understand the mechanisms underlying these effects and optimize treatment strategies for different fruit crops.



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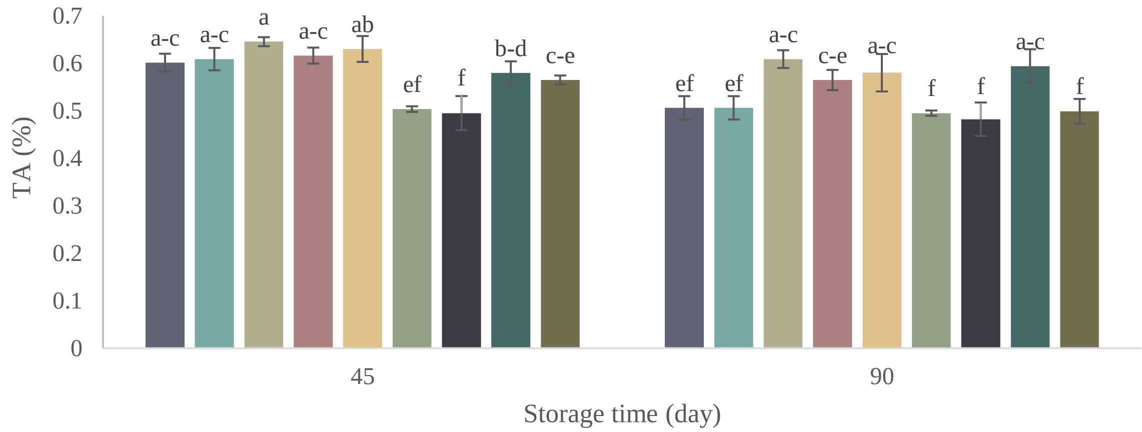

Fig. 3. The effect of different levels of melatonin spray and xanthan gum immersion on the content of total soluble solids (TSS) and TA of Orlando tangelo fruit.

Ascorbic acid

After 45 days of storage, melatonin 200 μM+ xanthan 0.1% and xanthan 0.1% showed the highest ascorbic acid content, which was significantly different from the control. At the end of the experiment, melatonin 100 μM+ xanthan 0.1% treatment showed a significantly higher level of ascorbic acid than the control, while the other treatments did not show a significant difference with the control (Fig. 4). Ascorbic acid is a potent antioxidant that protects fruits from the harmful effects of reactive oxygen species, as mentioned by Blokhina et al. (2003). Ascorbic acid acts as an antioxidant and undergoes decomposition upon reacting with oxygen. Consequently, the application of edible coatings that minimize oxygen contact has been found to effectively delay the oxidative degradation of ascorbic acid in fruits (Ayranci and Tunc 2004). Edible coatings are effective in delaying ascorbic acid degradation during storage. This is attributed to the formation of a protective layer that reduces the exposure of fruit skin to oxygen (Tigist et al., 2013). Studies have shown that pomegranate fruits from trees treated with melatonin contain more ascorbic acid compared to those from control trees. Furthermore, the coated samples exhibited less degradation of ascorbic acid than the uncoated samples, as reported by Kumar and Saini (2021). Furthermore, according to Xia et al.,

(2020) the application of 0.05 or 0.1 mM melatonin improved the concentration of ascorbic acid in cherry fruits. The study conducted by Shang et al. (2021) supports the positive effect of melatonin on ascorbic acid, although specific details have not been provided. These findings highlight the potential of melatonin treatment and edible coatings to preserve the ascorbic acid content in fruits by reducing oxidative degradation.

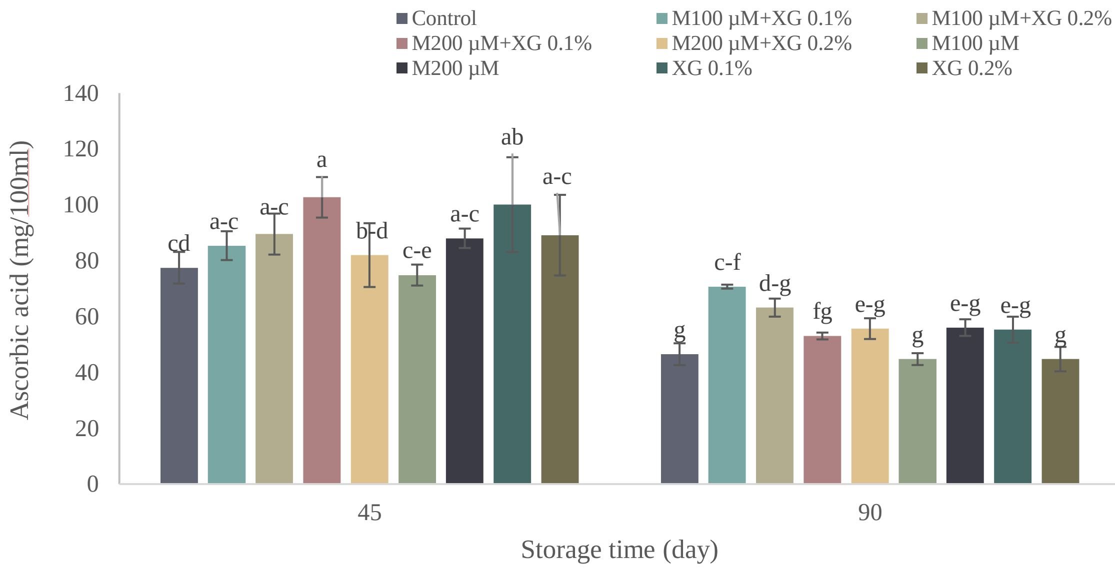

Fig 4- The effect of different levels of melatonin spray and xanthan gum immersion on the ascorbic acid of Orlando tangelo.

Total phenol, flavonoid and antioxidant capacity

After 45 days of storage, the phenolic content of the treated fruits was found to be higher than that of the control in most of the treatments. However, during the subsequent 90 days of storage, there was a significant decline in the phenolic content among all treatments, and no notable difference was observed between the control and the treated groups (Fig. 5a). Phenolic compounds are synthesized at a faster rate during the ripening process of fruits but tend to decrease as the fruit approaches senescence. These compounds play crucial roles in determining fruit quality and phytochemical levels (Shamloo et al. 2015). Phenolic compounds are known for their antioxidant properties and contribute to the sensory attributes, nutritional value, and potential health benefits of fruit. Phenolic compounds play a crucial role in maintaining the nutritional quality of fruits and vegetables by influencing their color, firmness, taste, and bitterness. These compounds represent a diverse group of secondary metabolites, with documented beneficial effects on human health. Phenolic compounds act as antioxidants and have various biological activities. The role of melatonin in modulating the total phenolic content of citrus fruits has been studied, and it has been found that melatonin treatment in oranges leads to the of total phenols during storage (Ma et al., 2021). For grapes, xanthan gum has been shown to preserve phenolic compounds by creating a protective coating around the grapes, which restricts oxygen supply and helps maintain phenolic content (Quoc et al., 2014). Furthermore, research has shown that melatonin treatment in pomegranate trees helps to maintain higher levels of phenolic compounds in fruits than in the control group throughout the storage period. Similarly, the application of melatonin has been reported to increase the concentration of phenolic compounds in grapes (Wang et al., 2020).

Based on the data presented in Fig. 5b, the flavonoid content during the storage period was consistently higher in the treated groups compared to the control. In particular, after 90 days of storage, the xanthan 0.2% treatment exhibited the highest flavonoid content among all the treatments. Flavonoids are a group of polyphenolic compounds that are released as part of the defense mechanisms of plants (Uckoo et al., 2015). These secondary metabolites enhance the antioxidant capabilities of fruits by eliminating free radicals. Additionally, a study conducted on Kinnow tangerines examined the impact of coatings on fruit quality. It was found that, , the total flavonoid content and juice content were better preserved than the control, indicating that the coatings helped maintain the flavonoid content of tangerines (Bajaj et al., 2024). These findings highlight the potential of melatonin and xanthan gum treatments to promote flavonoid accumulation in fruits.

Based on the data presented in Fig. 5c, fruit treated with melatonin 200 µM and 100 µM showed the highest antioxidant activity than other treatments in 45th day of storage. However, at the end of storage, melatonin 100 µM+ xanthan 0.1%, melatonin 200 µM+ xanthan 0.1% and + melatonin 100 µM showed higher antioxidant activity than the control. Melatonin improves the content of bioactive compounds and antioxidant activity in various fruit products (Wu et al., 2021). In grapes, coatings enriched with xanthan gum and acid phytochemicals were found to preserve the antioxidant and tissue properties of grapes during cold storage (Golly et al., 2019). Furthermore, studies have also shown that melatonin enhances the antioxidant activities of sweet cherry (Xia et al., 2020).

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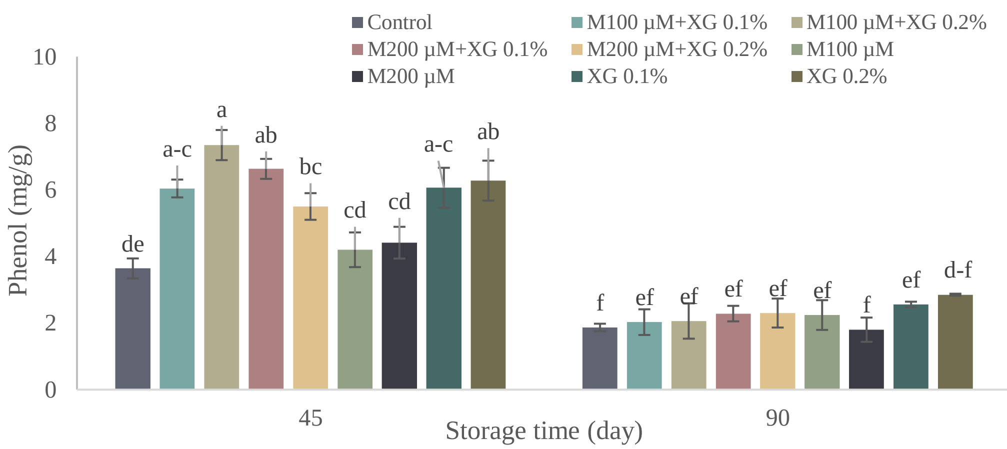

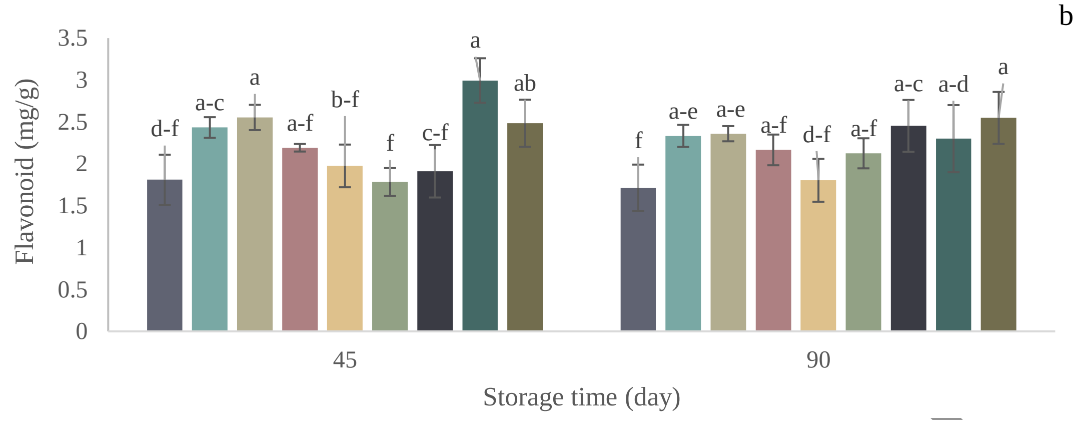

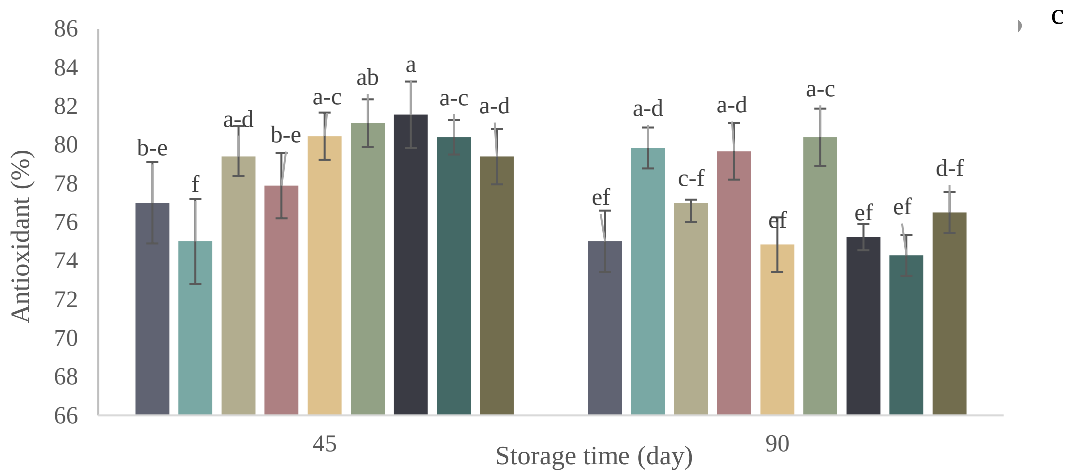

Fig 5- The effect of different levels of melatonin spray and xanthan gum immersion on the total phenol (a), flavonoid (b) and (c)Orlando Tangelo.

Conclusions

This experiment demonstrated that spray of melatonin at a concentration of 100 µM had significant effects on the characteristics of Orlando tangelo mandarin fruits. These effects included an increase in fruit weight and fruit pulp weight, as well as higher levels of ascorbic acid and increased fruit acidity, compared to the control. Throughout the experiment, the fruits treated with a combination of 200 µM melatonin and 0.1% xanthan gum exhibited the lowest weight loss. Furthermore, treatment with 100 µM melatonin and 0.1% xanthan gum showed the highest ascorbic acid content, while the treatments with 100 µM and 200 µM melatonin, both with 0.1% xanthan gum, exhibited the highest antioxidant activity. Overall, these findings suggest that preharvest spray of melatonin and the application of xanthan gum coating may be an effective strategy to maintain the quality of Orlando tangelo mandarin fruits during storage. These techniques have the potential to improve various characteristics such as weight, ascorbic acid content, acidity, and antioxidant activity.

Acknowledgments

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References:

- Arabia, A., Munne-Bosch, S., & Muñoz, P. (2022). Melatonin triggers tissue-specific changes in anthocyanin and hormonal contents during postharvest decay of Angeleno plums. *Plant Science*, *320*, 111287. https://doi.org/10.1016/j.plantsci.2022.111287

- Ayranci, E., & Tunc, S. (2004). The effect of edible coatings on water and vitamin C loss of apricots (*Armeniaca vulgaris* Lam.) and green peppers (*Capsicum annuum* L.). *Food chemistry*, *87*(3), 339-342. https://doi.org/10.1016/j.foodchem.2003.12.003

- Bajaj, K., Kumar, A., Gill, P. P. S., Jawandha, S. K., & Kaur, N. (2024). Xanthan gum coatings augmented with lemongrass oil preserve postharvest quality and antioxidant defence system of Kinnow fruit under low-temperature storage. *International Journal of Biological Macromolecules*, 129776. https://doi.org/10.1016/j.ijbiomac.2024.129776

- Bal, E. (2019). Physicochemical changes in 'Santa Rosa'plum fruit treated with melatonin during cold storage. *Journal of Food Measurement and Characterization*, *13*, 1713-1720. https://doi.org/10.1007/s11694-019-00088-6

- Baraiya, N. S., Ramana Rao, T. V., & Thakkar, V. R. (2016). Composite coating as a carrier of antioxidants improves the postharvest shelf life and quality of table grapes (Vitis vinifera L. var. Thompson Seedless). *Journal of Agricultural Science and Technology*, *18*(1), 93-107.

- Blokhina, O., Virolainen, E., & Fagerstedt, K. V. (2003). Antioxidants, oxidative damage and oxygen deprivation stress: a review. *Annals of botany*, *91*(2), 179-194. https://doi.org/10.1093/aob/mcf118

- Bor, J. Y., Chen, H. Y., & Yen, G. C. (2006). Evaluation of antioxidant activity and inhibitory effect on nitric oxide production of some common vegetables. *Journal of agricultural and food chemistry*, *54*(5), 1680-1686. https://doi.org/10.1021/jf0527448

- Brand-Williams W, Cuvelier M, Berset C (1995) Antioxidative activity of phenolic composition of commercial extracts of sage and rosemary. Lwt 28:25–30

- Carrión-Antolí, A., Martínez-Romero, D., Guillén, F., Zapata, P. J., Serrano, M., & Valero, D. (2022). Melatonin pre-harvest treatments leads to maintenance of sweet cherry quality during storage by increasing antioxidant systems. *Frontiers in Plant Science*, *13*, 863467. https://doi.org/10.3389/fpls.2022.863467

- Dong, F., & Wang, X. (2018). Guar gum and ginseng extract coatings maintain the quality of sweet cherry. *Lwt*, *89*, 117-122. https://doi.org/10.1016/j.lwt.2017.10.035

- Food and Agriculture Organization – FAO. 2020. International year of fruits and vegetables Retrieved from http://www.fao.org/documents/card/en/c/cb2395en/. Food and Drug Approvals-FD. 2020. https://www.fda.gov

- Gad, M. M., & Zagzog, O. A. (2017). Mixing xanthan gum and chitosan nano particles to form new coating for maintaining storage life and quality of elmamoura guava fruits. *International Journal of Current Microbiology and Applied Sciences*, *6*(11), 1582-1591. https://doi.org/10.20546/ijcmas.2017.611.190

Golly, M. K., Ma, H., Sarpong, F., Dotse, B. P., Oteng-Darko, P., & Dong, Y. (2019). Shelf-life extension of grape (Pinot noir) by xanthan gum enriched with ascorbic and citric acid during cold temperature storage. *Journal of food science and technology*, *56*, 4867-4878.

- Guccione, E., Allegra, A., Farina, V., Inglese, P., & Sortino, G. (2023). Use of xanthan gum and calcium ascorbate to prolong cv. Butirra pear slices shelf life during storage. *Advances in Horticultural Science*, *37*(1), 59-66. DOI: https://doi.org/10.36253/ahsc-13872

- Khatri, D., Panigrahi, J., Prajapati, A., & Bariya, H. (2020). Attributes of Aloe vera gel and chitosan treatments on the quality and biochemical traits of post-harvest tomatoes. *Scientia Horticulturae*, *259*, 108837. https://doi.org/10.1016/j.scienta.2019.108837

- Kittur, F. S., Saroja, N., Habibunnisa, & Tharanathan, R. (2001). Polysaccharide-based composite coating formulations for shelf-life extension of fresh banana and mango. *European food research and technology*, *213*, 306-311. https://doi.org/10.1007/s002170100363

- Kumar, A., & Saini, C. S. (2021). Edible composite bi-layer coating based on whey protein isolate, xanthan gum and clove oil for prolonging shelf life of tomatoes. *Measurement: Food*, *2*, 100005. https://doi.org/10.1016/j.meafoo.2021.100005

- Liu, J., Yue, R., Si, M., Wu, M., Cong, L., Zhai, R., ... & Xu, L. (2019). Effects of exogenous application of melatonin on quality and sugar metabolism in 'Zaosu'pear fruit. *Journal of Plant Growth Regulation*, *38*, 1161-1169. https://doi.org/10.1007/s00344-019-09921-0

- Li, X., Li, C., Sun, J., & Jackson, A. (2020). Dynamic changes of enzymes involved in sugar and organic acid level modification during blueberry fruit maturation. *Food chemistry*, *309*, 125617. https://doi.org/10.1016/j.foodchem.2019.125617

- López, L., Larrigaudière, C., Giné-Bordonaba, J., & Echeverria, G. (2023). Defining key parameters and predictive markers of 'Early Bigi'cherry consumer satisfaction by means of differential storage scenarios. *Postharvest Biology and Technology*, *195*, 112117. https://doi.org/10.1016/j.postharvbio.2022.112117

- Lorente-Mento, J. M., Guillén, F., Castillo, S., Martínez-Romero, D., Valverde, J. M., Valero, D., & Serrano, M. (2021). Melatonin treatment to pomegranate trees enhances fruit bioactive compounds and quality traits at harvest and during postharvest storage. *Antioxidants*, *10*(6), 820. https://doi.org/10.3390/antiox10060820

- Ma, Q., Lin, X., Wei, Q., Yang, X., Zhang, Y. N., & Chen, J. (2021). Melatonin treatment delays postharvest senescence and maintains the organoleptic quality of 'Newhall'navel orange (Citrus sinensis (L.) Osbeck) by inhibiting respiration and enhancing antioxidant capacity. *Scientia Horticulturae*, *286*, 110236. https://doi.org/10.1016/j.scienta.2021.110236

- Medina-Santamarina, J., Zapata, P. J., Valverde, J. M., Valero, D., Serrano, M., & Guillén, F. (2021). Melatonin treatment of apricot trees leads to maintenance of fruit quality attributes during storage at chilling and non-chilling temperatures. *Agronomy*, *11*(5), 917. https://doi.org/10.3390/agronomy11050917

- Omid, M., Khojastehnazhand, M., & Tabatabaeefar, A. (2010). Estimating volume and mass of citrus fruits by image processing technique. *Journal of food Engineering*, *100* (2), 315-321. https://doi.org/10.1016/j.jfoodeng.2010.04.015

- Ordonez, A. A. L., Gomez, J. D., & Vattuone, M. A. (2006). Antioxidant activities of Sechium edule (Jacq.) Swartz extracts. *Food chemistry*, *97*(3), 452-458. https://doi.org/10.1016/j.foodchem.2005.05.024

- Quoc L, Hoa D, Ngoc H, Phi T (2015) Effect of xanthan gum solution on the preservation of acerola (Malpighia glabra L.). Cercetari Agronomice in Moldova 48:89–97. https://doi.org/10.1515/cerce-2015-0045

- Rastegar, S., Khankahdani, H. H., & Rahimzadeh, M. (2020). Effects of melatonin treatment on the biochemical changes and antioxidant enzyme activity of mango fruit during storage. *Scientia Horticulturae*, *259*, 108835. https://doi.org/10.1016/j.scienta.2019.108835

-Rastegar, S., Aghaei Dargiri, S., & Mohammadi, M. (2024). Mitigating postharvest chilling injury in Orlando tangelo fruit: potential of melatonin and GABA in enhancing the antioxidant system. *Acta Physiologiae Plantarum*, *46*(3), 30. https://doi.org/10.1007/s11738-024-03653-9

- Ruelas-Chacon, X., Contreras-Esquivel, J. C., Montañez, J., Aguilera-Carbo, A. F., Reyes-Vega, M. L., Peralta-Rodriguez, R. D., & Sanchéz-Brambila, G. (2017). Guar gum as an edible coating for enhancing shelf-life and improving postharvest quality of roma tomato (Solanum lycopersicum L.). *Journal of Food Quality*, *2017*. https://doi.org/10.1155/2017/8608304

- Saini, R. K., Ranjit, A., Sharma, K., Prasad, P., Shang, X., Gowda, K. G. M., & Keum, Y. S. (2022). Bioactive compounds of citrus fruits: A review of composition and health benefits of carotenoids, flavonoids, limonoids, and terpenes. *Antioxidants*, *11*(2), 239. https://doi.org/10.3390/antiox11020239

- Salehi, F. (2020). Edible coating of fruits and vegetables using natural gums: A review. *International Journal of Fruit Science*, *20*(sup2), S570-S589. https://doi.org/10.1080/15538362.2020.1746730

- Shahid, M. N., & Abbasi, N. A. (2011). Effect of bee wax coatings on physiological changes in fruits of sweet orange CV."blood red". *Sarhad Journal of Agriculture*, *27*(3), 385-394.

- Shamloo, M.M., Sharifani, M., Daraei Garmakhany, A., & Seifi, E. (2015). Alternation of secondary metabolites and quality attributes in Valencia Orange fruit (*Citrus sinensis*) as influenced by storage period and edible covers. *Journal of Food Science and Technology*, *52*(4), 1936-1947. https://doi.org/10.1007%2Fs13197-013-1207-4

- Shang, F., Liu, R., Wu, W., Han, Y., Fang, X., Chen, H., & Gao, H. (2021). Effects of melatonin on the components, quality and antioxidant activities of blueberry fruits. *Lwt*, *147*, 111582. https://doi.org/10.1016/j.lwt.2021.111582

- TRAORE, M., DIARRA, S., SISSOKO, S., DIAWARA, M. O., TRAORE, M., TRAORE, B. M., .. & SIDIBE, A. (2023). Determinism of some phytohormones and potassium nitrate on the vegetative growth of Tangelo (Citrus x tangelo) in the Sudano-Sahelian area of Mali. *GSC Biological and Pharmaceutical Sciences*, *23*(1), 160-167. https://doi.org/10.30574/gscbps.2023.23.1.0147

- Tigist, M., Workneh, T. S., & Woldetsadik, K. (2013). Effects of variety on the quality of tomato stored under ambient conditions. *Journal of food science and technology*, *50*, 477-486. https://doi.org/10.1007/s13197-011-0378-0

- Tripathi, A. D., Sharma, R., Agarwal, A., & Haleem, D. R. (2021). Nanoemulsions based edible coatings with potential food applications. *International Journal of Biobased Plastics*, *3*(1), 112-125. https://doi.org/10.1080/24759651.2021.1875615

- Uckoo, R. M., Jayaprakasha, G.K., & Patil, B.S. (2015). Phytochemical analysis of organic and conventionally cultivated Meyer limes (*Citrus meyeri* Tan.) during refrigerated storage. *Journal of Food Composition and Analysis*, *42*, 63-70. http://dx.doi.org/10.1016/j.jfca.2015.01.009

- Verde, A., Míguez, J. M., & Gallardo, M. (2022). Role of melatonin in apple fruit during growth and ripening: possible interaction with ethylene. *Plants*, *11*(5), 688. https://doi.org/10.3390/plants11050688

- Vignesh, R. M., & Nair, B. R. (2019). Improvement of shelf life quality of tomatoes using a novel edible coating formulation. *Plant Science Today*, *6*(2), 84-90. https://doi.org/10.14719/pst.2019.6.2.443

- Wang, L., Luo, Z., Yang, M., Li, D., Qi, M., Xu, Y. ... & Li, L. I. (2020). Role of exogenous melatonin in table grapes: First evidence on contribution to the phenolics-oriented response. *Food chemistry*, *329*, 127155. https://doi.org/10.1016/j.foodchem.2020.127155

- Wang, F., Zhang, X., Yang, Q., & Zhao, Q. (2019). Exogenous melatonin delays postharvest fruit senescence and maintains the quality of sweet cherries. *Food chemistry*, *301*, 125311. https://doi.org/10.1016/j.foodchem.2019.125311

- Wu, X., Ren, J., Huang, X., Zheng, X., Tian, Y., Shi, L.,... & Li, Z. (2021). Melatonin: Biosynthesis, content, and function in horticultural plants and potential application. *Scientia Horticulturae*, *288*, 110392. https://doi.org/10.1016/j.scienta.2021.110392

- Xia, H., Shen, Y., Shen, T., Wang, X., Zhang, X., Hu, P., ... & Lv, X. (2020). Melatonin accumulation in sweet cherry and its influence on fruit quality and antioxidant properties. *Molecules*, *25*(3), 753. https://doi.org/10.3390/molecules25030753

- Xu, L. L., Yue, Q. Y., Bian, F. E., Zhai, H., & Yao, Y. X. (2017). Effects of melatonin treatment on grape berry ripening and contents of ethylene and ABA. *Acta Phytophysiol Sin*, *53*, 2181-2188. https://doi.org/10.3389/fpls.2017.01426

- Zhao, L., Yan, S., Wang, Y., Xu, G., & Zhao, D. (2023). Evaluation of the Effect of Preharvest Melatonin Spraying on Fruit Quality of 'Yuluxiang'Pear Based on Principal Component Analysis. *Foods*, *12*(18), 3507. https://doi.org/10.3390/foods12183507