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Maintaining quality of Lisbon lemon (*Citrus limon*) in cold storage using natural elicitors

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ABSTRACT

Purpose: Lemon (Citrus limon) is a highly important citrus species worldwide. However, its semi-tropical nature makes it susceptible to chilling, extensive research on postharvest treatments to preserve its quality under low temperatures. Research Method: The treatments included 500 µM melatonin (M), 50 µM methyl jasmonate (J), and 5 mM gamma-aminobutyric acid (GABA). Lemons stored at 3 ± 1°C and a relative humidity of 85-95% for 100 days plus 5-day shelf life. Findings: The results revealed that most of the experimental treatments, except for the combination of GABA + J and GABA + J + M, significantly reduced fruit weight loss. Notably, the melatonin treatment showed a 22.7% lower weight loss compared to the control fruits. Furthermore, the melatonin treatment exhibited the highest fruit firmness (49 N), while the control treatment had the lowest (36.3 N). Regarding the quality parameters, individual treatments, and the GABA + M treatment resulted in significantly higher total soluble solids (TSS) and a lower TSS/TA compared to the control at the end of the storage period. Except for the M + GABA and M + J treatments, all other treatments showed higher ascorbic acid content compared to the control. Additionally, the melatonin treatment showed significant differences in various color indices compared to the control. Research limitations: There was no limitation. Originality/Value: Overall, fruits treated with M, J, GABA, and GABA + M demonstrated higher marketability compared to the control and other treatments. Consequently, it is recommended to utilize these treatments individually rather than in combination to maintain the quality of lemon fruits.

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INTRODUCTION

Lemon fruit (Citrus limon cv. Lisbon) is rich in nutrients and bioactive compounds such as citric acid, ascorbic acid, minerals and rich in natural antioxidant compounds such as phenols and flavonoids and antioxidant properties (Serna-Escolano et al., 2021). The quality of lemon depends on the physiological and biochemical changes that occur during its storage conditions and duration (Mukhim et al., 2015). Cold storage facilities play a crucial role in expediting the desired decrease in temperature, thereby minimizing postharvest losses, and safeguarding the integrity and volume of perishable agricultural produce. By upholding appropriate cold storage conditions, the nutritional and sensory attributes of freshly harvested fruits and vegetables can be effectively preserved, thereby affording consumers with a diverse array of processing prospects (Makule et al., 2022). However, some tropical and semi-tropical products, especially lemons, are very sensitive to low temperature, and their long-term storage in cold storage leads to a decrease in some quality characteristics such as taste, properties affecting fruit juice and bioactive compounds such as ascorbic acid (Rapisarda et al., 2001; Baloyi et al., 2023) and the marketability of the product (Sun et al., 2019). The occurrence of chilling symptoms is due to lipid peroxidation along with a decrease in the activity of antioxidant mechanisms because of excessive production of ROS and because of oxidative stress. In general, all kinds of subtropical fruits, such as lemon, have developed protective mechanisms against oxidative stress. For example, fruits have developed efficient systems (enzymatic antioxidants, liposoluble or membrane-bound antioxidants, and water-soluble antioxidants) to scavenge ROS (Habibi et al., 2020).

In this regard, to increase the tolerance of citrus fruits against chilling during storage, various compounds such as salicylic acid and edible coatings (Rasouli et al., 2019), methyl jasmonate and methyl salicylate (Habibi et al., 2019) and nitric Oxide (Ghorbani et al., 2018) has been studied. These compounds activate new signaling pathways to deal with stress. The studies conducted are mostly on orange and grapefruit fruits and the use of natural compounds such as melatonin, methyl jasmonate and GABA on Lisbon lemon is very limited.

Melatonin is a natural indoleamine and an effective antioxidant that directly destroys ROS. Recently, strong evidence has been obtained that shows that plants produce and accumulate melatonin when exposed to adverse environmental conditions, and the use of exogenous melatonin helps to increase tolerance to stress (Jannatizadeh, 2019). Application of melatonin for 'Guifei' mango effectively delayed the change in ripening parameters including firmness, color, total soluble solids content and titratable acidity (TA) by inhibiting ethylene and ABA biosynthesis (Liu et al., 2020).

Methyl jasmonate, known for its ability to enhance the quality of fresh fruits both preand post-harvest, achieves this by promoting the production of secondary metabolites and increasing antioxidant activity. These effects contribute to an overall improvement in fruit quality. Moreover, Methyl jasmonate application can also enhance plant resistance against storage-related problems like chilling injuries and pathogen attacks, effectively preserving the quality of the fruits (Akan et al., 2019). In another study, methyl jasmonate treatment on blood orange delayed the changes in fruit firmness, TA, antioxidant activity and ascorbic acid, and the treated fruit contained higher compounds and showed higher cold resistance (Habibi et al., 2020). MeJA treatments significantly retarded weight loss in blueberries during storage. While the values of L*, chroma, and soluble solids content in MeJA-treated blueberries were lower (Y1lmaz, 2024).

GABA is a four-carbon, non-protein amino acid. This compound in plants is known as a metabolite in the field of response to living and non-living stresses that can quickly accumulate in response to stresses and includes the defense system against them (Ramesh et



al., 2015). Research findings have demonstrated that the utilization of GABA on olives led to improved quality during periods of cold storage in comparison to untreated olives. This improvement was attributed to the increased functioning of antioxidant enzymes in the olives that received GABA treatment (Fan et al., 2024).

In recent years, there has been a growing interest in employing environmentally friendly methods to extend the shelf life of agricultural products without compromising human health. The innovation of the article lies in evaluating, for the first time, the effects of melatonin, methyl jasmonate, gibberellic acid, and their combination on the quality of stored lemon fruit in cold storage. Prior to this research, there have been no reports on the use of these compounds and their impact on the quality of lemon fruit during storage. This study aims to explore the potential benefits and effects of these substances on the characteristics of stored Lisbon lemon fruit at a temperature of $3\pm1^{\circ}$ C. By investigating the effects of natural materials, this research aims to contribute to the development of sustainable and safe practices in the agricultural industry.

MATERIALS AND METHODS

Lemon fruits (*Citrus limon* cv. Lisbon) were harvested by hand from a commercial orchard in Juyom city, Fars province, and were immediately transported to the physiology laboratory for treatment and trait evaluation. First, uniform fruits were selected in terms of color, shape, and size, and non-uniform and damaged samples were removed, then they were immersed in 0.05% sodium hypochlorite solution for one minute to remove any contamination and washed immediately. Then the fruits were subjected to immersion treatment (10 minutes) in melatonin 500 μ M, methyl jasmonate 50 μ M and GABA 5 mM. Control fruits were immersed in distilled water for 10 min. Each treatment included 3 repetitions and each repetition included 3 fruits. After applying the treatment, the fruits were kept at ambient temperature and after drying, they were transferred to the storage (temperature 3±1°C and relative humidity 85-90%). At intervals of 30, 70, and 100 days during the cold storage period, the samples were assessed to measure various parameters (Siboza et al., 2014).

Characteristics under investigation

Weight loss index

To measure the percentage of weight loss (WL), the fruits in each experimental unit were weighed with an accuracy of 0.01 grams as the initial weight after treatment and before storage. Then, at each sampling time, they were re-weighed as the final weight, and finally, according to the amount of initial weight and secondary weight, the percentage of weight loss was calculated according to the following equation: The percentage of weight loss reported with the following equation (1) (Habibi et al., 2020):

Weight loss (%) = ((Initial weight – Final weight) / (Initial weight))
$$\times 100$$
 (1)

Firmness index

The hardness of each fruit was determined with a hardness tester equipped with a probe with a diameter of 3.5 mm. The prop of the device compressed 10% of the equatorial diameter of the fruit and stopped after splitting the skin of the fruit and the data was reported in Newton (N).

Taste index, Total soluble solids, Titratable acidity

Total soluble solids (TSS) in water samples were determined using a digital hand-held refractometer (Atago PAL-1, Tokyo, Japan) and reported as °Brix.



Titratable acidity (TA) was determined by manual titration of lemon juice with 0.1 N NaOH to pH 8.2 as an endpoint with a pH meter (HANA, Romani) as an indicator and expressed as a percentage of citric acid equivalent (2).

TA (% citric acid) = $0.0064 \times \text{titre}$ (NaOH) mL×100/ 10 mL juice (2)

The taste index (TSS/TA) was expressed as the ratio of TSS to TA (Habibi et al., 2020).

Ascorbic acid content

Ascorbic acid (AA) content in fruit juice was measured using 2,6-dichlorophenol indophenol method. 50 μ l of lemon juice was added to 5 ml of 1% metaphosphoric acid and vortexed for 20 seconds, and then 500 μ l of the vortexed solution was combined with 4.5 ml of indophenol and vortexed again for 20 seconds. Then it was read using a spectrophotometer at a wavelength of 510 nm and reported as mg/100g FW: (AOAC, 2000; Rey et al., 2020).

Fruit color index

The color characteristics of lemon fruit were monitored using a colorimeter (Minolta CR400, Japan), with the CIE L*, a* and b* system proposed by the International Commission on Eclairage (CIE). To measure the skin color of the fruit at each measurement stage, three fruits were selected from each repetition and the readings were taken from three different points on the fruit. The L^* value indicates darkness or lightness, the a^* value is green (-) or red (+), and the b^* value is yellow (+) or blue (-) (Sun et al., 2019).

Marketability

The marketability of lemons was evaluated by 10 evaluators based on the appearance indicators of fruit color, degree of skin freshness, lack of stains from frostbite, etc. using grading and scoring. So that zero has the lowest marketability and 10 has the highest marketability.

Experimental design and data analysis

The experiment was conducted as a factorial in the form of a completely randomized design with three replications (n=3). Statistical analysis of data was done using (9.1) SAS. The mean comparison was analyzed with Duncan's multiple range tests at the 5% probability level. Graphs were also drawn using Excel software.

RESULTS

Weight loss index

The results obtained from the mean comparison showed that the weight loss index increased over time, but this increase was less in the melatonin treatment than in the other treatments and the control. All treatments except GABA + methyl jasmonate and the combination of three treatments melatonin + GABA + methyl jasmonate showed a significant decrease in all three storage times compared to the control and caused a decrease in the percentage of weight loss and juice loss (Fig. 1).



Fig. 1. The effect of melatonin (M), methyl jasmonate (J), GABA (G), melatonin + GABA (M+G), methyl jasmonate + GABA (G+J), melatonin + methyl jasmonate (M+J) and melatonin + methyl jasmonate + GABA (M+J+G) treatments on the weight loss percentage of Lisbon lemons during the cold storage period including 30+5, 70+5, and 100+5 days stored at $3\pm1^{\circ}$ C. Similar letters indicate non-significant difference at the 5% probability level using Duncan's test. The vertical index above the columns represents the standard error.



Fig. 2. The effect of melatonin (M), methyl jasmonate (J), GABA (G), melatonin + GABA (M+G), methyl jasmonate + GABA (G+J), melatonin + methyl jasmonate (M+J) and melatonin + methyl jasmonate + GABA (M+J+G) treatments on the firmness of Lisbon lemons during the period of storage in cold storage including 30+5, 70+5 and 100+5 days stored at $3\pm1^{\circ}$ C. Similar letters indicate non-significant difference at the 5% probability level using Duncan's test. The vertical index above the columns represents the standard error.

Firmness index

Comparison of means shows that by as storage period increased, the firmness gradually decreased. According to Figure 2, after 100 days of storage, the fruits treated with melatonin, methyl jasmonate and GABA alone showed the highest level of firmness and the control treatment showed the lowest level of firmness, respectively.



Taste index, Total soluble solids, Titratable acidity

Comparison of means indicates that the soluble solids slightly increased with the progress of storage time. According to Figure 3, this increase was the highest in the control fruits, and on the other hand, the melatonin-treated fruits showed the lowest soluble solids in all three storage times, but no significant difference was observed between the other treatments. In the last stage, melatonin, GABA, methyl jasmonate and the combination of melatonin and GABA showed a significant decrease compared to the control. During the storage period, there was a notable decrease in titratable acidity content in the samples. Specifically, after 70 days of storage, the control samples exhibited a lower acid content compared to the samples treated with melatonin and methyl jasmonate. However, it is worth noting that by the end of the storage period, no statistically significant difference in titratable acidity content was observed between the control samples and the treated samples (Fig. 4).

The comparison of means indicates that the taste index has gradually increased with the progress of the shelf life of the fruits in the cold storage. At the end of the experiment, except GABA + jasmonate, melatonin+ jasmonate and melatonin + GABA + jasmonate treatments, other treatments showed less TSS than the control (Fig. 5).

Ascorbic acid content

The results of means comparison show that as the storage life of fruits increases from 30 to 100 days, the amount of ascorbic acid decreases. This reduction in the control treatments was very noticeable and had a significant difference compared to the rest of the treatments except the combination of melatonin with GABA and melatonin with methyl jasmonate (Fig. 6).



Fig. 3. Effect of melatonin (M), methyl jasmonate (J), GABA (G), melatonin + GABA (M+G), methyl jasmonate + GABA (G+J), melatonin + methyl jasmonate (M+J) and melatonin + methyl jasmonate + GABA (M+J+G) treatments on the soluble solids of the whole Lisbon lemon during the cold storage period including 30+5, 70+5 and 100+5 days stored at $3\pm1^{\circ}$ C. Similar letters indicate non-significant difference at the 5% probability level using Duncan's test. The vertical index above the columns represents the standard error.



Fig. 4. The effect of melatonin (M), methyl jasmonate (J), GABA (G), melatonin + GABA (M+G), methyl jasmonate + GABA (G+J), melatonin + methyl jasmonate (M+J) and melatonin + methyl jasmonate + GABA (M+J+G) treatments on the percentage of titratable acidity of Lisbon lemon during the period of storage in cold storage including 30+5, 70+5, and 100+5 days at $3\pm1^{\circ}$ C. Similar letters indicate non-significant difference at the 5% probability level using Duncan's test. The vertical index above the columns represents the standard error.



Fig. 5. The effect of melatonin (M), methyl jasmonate (J), GABA (G), melatonin + GABA (M+G), methyl jasmonate + GABA (G+J), melatonin + methyl jasmonate (M+J) and melatonin + methyl jasmonate + GABA (M+J+G) treatments on the taste index of Lisbon lemons during the period of storage in cold storage including 30+5, 70+5 and 100+5 days at a temperature of $3\pm1^{\circ}$ C. Similar letters indicate non-significant difference at the 5% probability level using Duncan's test. The vertical index above the columns represents the standard error.



Fig. 6. The effect of melatonin (M), methyl jasmonate (J), GABA (G), melatonin + GABA (M+G), methyl jasmonate + GABA (G+J), melatonin + methyl jasmonate (M+J) and melatonin + methyl jasmonate + GABA (M+J+G) treatments on the amount of ascorbic acid in Lisbon lemons during the period of storage in cold storage including 30+5, 70+5 and 100+5 days at a temperature of 3 ± 1 °C. Similar letters indicate non-significant difference at the 5% probability level using Duncan's test. The vertical index above the columns represents the standard error.

Fruit color index

According to the results of means comparison, all fruit color traits (L*, a*, b*) were significantly ($P \le 0.05$) affected by treatments and storage periods in cold storage. According to Figure 7a, L* value increased over time during fruit storage in cold storage. This increase was much higher in control fruits. The fruits stored for 30 days showed significantly the lowest amount of L* compared to the fruits stored for 70 days and 100 days. The amount of a* in cold storage fruits treated with melatonin was significantly lower than the control fruits and the rest of the treatments. These results indicate that melatonin treatment has been able to significantly prevent the color change and chlorophyll degradation of lemons (Fig. 7b).

According to the obtained results, cold storage fruits treated with melatonin significantly had the lowest b* value compared to control fruits. Also, fruits showed a significant increase in b* values from 30 days to 100 days of cold storage. The lowest amount of b* was observed in fruits treated with melatonin after 30, 70 and 100 days of cold storage and the highest amount of b* was observed in control fruits (Fig. 7c).



Fig. 7. The effect of melatonin (M), methyl jasmonate (J), GABA (G), melatonin + GABA (M+G), methyl jasmonate + GABA (G+J), melatonin + methyl jasmonate (M+J) and melatonin + methyl jasmonate + GABA (M+J+G) treatments on fruit color properties (A: L*, B: a*, and C: b*) of Lisbon lemon during the cold storage period including 30+5, 70+5 and 100+5 days at a temperature of 3 ± 1 °C. Similar letters indicate non-significant difference at the 5% probability level using Duncan's test. The vertical index above the columns represents the standard error.

Marketability

At the end of the storage period, the fruits treated with melatonin, jasmonate and GABA showed the highest level of appearance quality, freshness and marketability compared to the control fruits (Fig. 8).



Fig. 8. The effect of melatonin (M), methyljasmonate (J), GABA (G), melatonin + GABA (M+G), methyljasmonate + GABA (G+J), melatonin + methyljasmonate (M+J) and melatonin + methyl jasmonate + GABA (M+J+G) treatments on the marketability of Lisbon lemon fruit during the cold storage period including 30+5, 70+5 and 100+5 days stored at $3\pm1^{\circ}$ C.



DISCUSSION

According to the report, all treatments demonstrated a reduction in weight loss compared to the control fruit during the 100 ± 5 days of storage. After harvesting, the water content of the product gradually decreases due to transpiration and the respiration process (Siboza et al., 2014). This leads to wilting, loss of quality, and other undesirable changes in the product, ultimately reducing its economic value. The decrease in weight loss observed in fruits treated with methyl jasmonate and GABA can be attributed to their effect on maintaining membrane integrity at low temperatures, as reported in previous studies on pomegranate and blood orange (Habibi et al., 2019; Sayyari et al., 2011). However, the melatonin treatment exhibited the lowest percentage of weight loss compared to the other treatments. Melatonin may reduce weight loss by modulating water transport across the cuticle and plasma membrane, as well as by reducing respiration. Other findings suggest that melatonin treatment induces the expression of cuticle formation genes (CER1 and GPAT4/8) while downregulating aquaporins PIP1;4 and PIP2;7 in treated sweet cherry fruits, indicating its potential role in modulating, or reducing water loss. The thickness and specific composition of cuticle waxes and cutin are known to play a role in regulating the permeability of the cuticle against water movement (Lara et al., 2014). Additionally, as noted by Hui et al. (2016), the preservation of membrane integrity and reduced membrane permeability in melatonin-treated peach fruits may be attributed to the higher accumulation of endogenous melatonin, indicating its capacity to scavenge reactive oxygen species (ROS) and indirectly maintain the stability of the cell membrane. Studies have demonstrated that the application of melatonin treatment results in a significant reduction in the rate of weight loss during postharvest storage of 'Newhall' navel orange fruit (Ma et al., 2021). It has been demonstrated that GABA reduces metabolic activity in fruit tissue, thereby preventing water loss and weight loss. This effect is achieved by decreasing the activity of antioxidant enzymes and minimizing stress on the fruit (Asgarian et al., 2022). It has been demonstrated that the application of methyljasmonate treatment resulted in the lowest weight loss (3.2%) compared to the control group (8.4%) during the cold storage period of 'Kinnow' mandarins (Baswal et al., 2020).

Fruit firmness is an essential characteristic that not only affects customers' assessment of the quality of newly harvested lemons but also the determination of their ability to be preserved during postharvest operations. Melatonin inhibits the process of cell wall disintegration through the suppression of enzymatic activities and gene expressions that are associated with cell wall degradation, namely pectin methylesterase (PME), polygalacturonase (PG), and cellulase (Cel). Consequently, this leads to a postponement of fruit softening and senescence (Liao et al., 2024). The investigation conducted by Rastegar et al. (2020) illuminates the beneficial impact of melatonin in maintaining the firmness of mango fruit throughout the storage period. Melatonin enhances the activity of antioxidant enzymes, reduces oxidative damage, and maintains the energy status of postharvest fruit, which contributes to maintaining firmness. Melatonin interacts with reactive oxygen species (ROS) and nitric oxide (NO), which enhances the antioxidant and defense systems in postharvest fruit, leading to the preservation of firmness. The multiple functions of melatonin, including its antioxidant properties and interaction with signaling molecules, play a role in delaying the softening and maintaining the firmness of postharvest fruit (Ze et al., 2021; Liao et al., 2024).

Research findings have indicated that treating carambola fruit with GABA has a beneficial impact on preserving firmness, even when subjected to chilling and non-chilling stresses. It is widely recognized that the decline in postharvest fruit firmness is primarily linked to the ripening stage's respiration process (Ngaffo Mekontso et al., 2021). Studies have

shown that the application of methyl jasmonate has proven effective in slowing down the activities of pectin methylesterase and cellulase enzymes, which are directly involved in fruit softening. As a result, 'Kinnow' mandarins treated fruit was able to maintain its firmness for a duration of up to 75 days during cold storage (Baswal et al., 2020).

Total soluble solids (TSS) in fruits include sugars and a small percentage of amino acids, organic acids, vitamins, and minerals. TSS is a valuable quality parameter related to consumer acceptance and has a significant impact on fruit taste and is considered one of the chemical indicators (Liu et al., 2020). The soluble solids increase as the fruit ripens. The reason for this gradual increase in Brix is the gradual decrease in fruit juice that occurs with the passage of time and during the storage period, which causes a decrease in solids dissolved in water and as a result, an increase in Brix. In other words, the more water the fruit has, the lower the weight loss, the lower the Brix. As previously reported, postharvest treatments delayed TSS increases during cold storage, possibly due to delays in ripening and aging processes. Melatonin treatment effectively delays the natural increase of TSS during storage. Changes in TSS may be related to changes in the shape of sugars during storage.

Titratable acidity decreased in all treatments during the storage period, although in general, the control fruit had lower titratable acidity values than the control throughout the entire storage period. The titratable acidity is related to the organic acids, which strongly affects the quality of the fruit. Its reduction during cold storage is related to using them as the main respiratory substrates for energy production during storage (Habibi et al., 2019; Liu et al., 2020). The taste index (TSS/TA) or the ratio of soluble solids to titratable acid is directly related to these two factors, and changes in these two factors affect the value of the taste index. This index is one of the important factors in determining fruit ripening and its quality and marketability (Hui et al., 2016) and its increase during cold storage was due to the decrease in organic acids and slight increase in sugars. However, all treatments significantly delayed the increase in flavor index and showed a possible effect on reducing the post-harvest ripening process.

Antioxidant compounds such as ascorbic acid are an important parameter in discussing the quality of fruits and vegetables (Khorshidi et al. 2011). It is worth mentioning that the higher concentration of ascorbic acid in the treated fruit causes higher antioxidant activity and beneficial health effects of lemon fruit consumption (Siboza et al., 2014). Antioxidant systems play an important role in quenching ROS and maintaining cellular redox homeostasis, thus modulating aging processes in plants (Jimenez et al., 2002). The treated fruits had higher ascorbic acid than the control fruit, and the decrease of ascorbic acid in the control samples was probably related to the aging of the fruit after long-term storage. These compounds maintained a higher content of ascorbic acid probably due to the reduction of ascorbic acid oxidase activity and delayed ripening as well as the aging process (Habibi et al., 2020). Postharvest application of methyl jasmonate maintained higher ascorbic acid content in pomegranate (Sayyari et al., 2011). Ascorbic acid is very susceptible to degradation due to oxidation during cold storage. Low temperature can increase the accumulation of free radicals. Ascorbic acid is a major and prominent non-enzymatic antioxidant in citrus fruits that has the potential to eliminate free radicals in cells (Huang et al., 2008). Therefore, the main reason for the reduction of ascorbic acid can be attributed to the inhibition of ROS by the non-enzymatic system as an electron donor to neutralize free radicals during storage at low temperature. In addition, the reduction of ascorbic acid is associated with fruit ripening after long-term storage (Habibi et al., 2019).

The assessment of fruit quality relies heavily on the color of the fruit, as it communicates the visual attractiveness of the fruit and has a significant impact on consumer preferences. Commercial maturity indexes in the citrus industry are usually based on peel coloration,



which is an external characteristic that defines fruit quality (Lado et al., 2018). The alteration of fruit color can occur during the storage process, and this alteration is influenced by various factors such as the duration of storage, the temperature within the cold storage facility, as well as any supplementary treatments applied. Specifically, citrus fruits display modifications in color that can be attributed to the breakdown of chlorophyll pigments and the buildup of carotenoids in flavonoids as they undergo the ripening process (Vidal et al., 2013). Mature citrus fruits are prepared with chromoplasts which possess the capacity to accumulate substantial quantities of carotenoids. These carotenoids, functioning as photoprotective agents, perform a vital function in safeguarding the integrity of the fruit's membranes, thus preventing any potential damage (Serna-Escolano et al., 2021). Research has demonstrated that the utilization of melatonin can impede the deterioration of the* indicator, which signifies the redness aspect of color, leading to elevated values in comparison to the control fruit. Melatonin treatment was found to delay the color changes in yellow-fleshed peach fruit compared to the control group during the storage period. This suggests that melatonin treatment can help maintain the original color of yellow-fleshed peaches for a longer duration, potentially preserving their visual appeal and quality (Wu et al., 2023). Melatonin treatment can have a positive effect on color parameters in postharvest fruit by delaying ripening, senescence, and deterioration, and by enhancing cold resistance (Ze et al., 2021). It has been shown that GABA treatment can effectively preserve the original color of the treated material for a longer period, potentially enhancing its visual appeal and quality. GABA application protects thylakoid membranes, chloroplast envelopes, and reduces swelling, thereby alleviating chlorophyll degradation (Fan et al., 2024). In a study on mango fruit treated with melatonin, it was found that the treatment inhibited pericarp discoloration during storage, possibly due to enhanced membrane integrity (Liu et al., 2020). Furthermore, the application of methyl jasmonate has shown to effectively maintain the color of the fruit. Similar observations have been documented, indicating that this treatment delays the browning of pomegranate skin and ensures the preservation of its overall appearance quality and color (Lorente-Mento, 2023). The treatments exert their effects by delaying metabolic processes, thereby slowing down the color change process.

Freshness has been documented as a primary factor influencing consumer selection of various fruits, such as citrus. Moreover, the visual aspect of fruits significantly contributes to consumers' perception of their freshness (Tarancón et al., 2021). According to the results, the appearance and marketability of the treated fruits were better than the control. The appearance quality and marketability are related to many factors such as the color and water content of the tissue, which damage caused by low temperature and water loss, respectively, by changing the color and drying of the peel during the storage period, can cause a decrease in the appearance quality. In addition, the decrease in appearance quality can be related to the oxidation of antioxidant compounds such as phenols due to the higher activity of polyphenol oxidase and peroxidase and the creation of brown pigment. The reduction of these phenolic compounds causes a change in the brightness of the skin (Habibi et al., 2020). Previous reports have indicated that melatonin treatment enhances the appearance of kiwiberries during storage by increasing the accumulation of total chlorophyll and carotenoids, thereby improving the flesh color of the fruit (Zhang et al., 2023). Several research groups have provided evidence that methyl jasmonate effectively improves color appearance. This positive impact is largely attributed to the ability of methyl jasmonate to enhance the antioxidant capacity of postharvest fruits. Furthermore, the improved color appearance can be attributed to the increased content of anthocyanins, which effectively mitigate skin browning (Wang et al., 2021).



CONCLUSION

In conclusion, this study investigated the effects of various postharvest treatments on lemons stored under low temperatures. The results demonstrated that most of the treatments, except for specific combinations, effectively reduced fruit weight loss. The melatonin treatment showed a significant decrease in weight loss compared to the control. Additionally, the melatonin treatment exhibited the highest fruit firmness. Regarding quality parameters, individual treatments, and the GABA + M treatment resulted in higher total soluble solids and a lower TSS/TA ratio compared to the control. Except for specific combinations, all treatments showed increased ascorbic acid content compared to the control. The melatonin treatment also showed notable differences in color indices. Overall, fruits treated with melatonin, methyl jasmonate, GABA, and the combination of GABA + M demonstrated higher marketability. Therefore, it is recommended to use these treatments individually to maintain the quality of lemon fruits. These findings provide valuable insights for the agricultural industry, suggesting the potential use of these natural compounds to enhance the shelf life and market value of lemons. Future research could explore optimization techniques for the application of these treatments and investigate their efficacy on other fruit varieties.

Conflict of interest

The authors declare that there is no conflict of interest.

REFERENCES

- Akan, S., Tuna Gunes, N., & Yanmaz, R. (2019). Methyl jasmonate and low temperature can help for keeping some physicochemical quality parameters in garlic (*Allium sativum* L.) cloves. *Food Chemistry*, 270, 546–553. https://doi.org/https://doi.org/10.1016/j.foodchem.2018.07.085
- AOAC. (2000). Vitamins and other nutrients, official methods of analysis (17th ed.). Washington, D.C.: AOAC International16–20.
- Asgarian, Z.S., Karimi, R., Ghabooli, M., & Maleki, M. (2022). Biochemical changes and quality characterization of cold-stored 'Sahebi'grape in response to postharvest application of GABA. *Food Chemistry*, *373*, 131401. https://doi.org/10.1016/j.foodchem.2021.131401
- Baloyi, R. G., Mafeo, T. P., & Mathaba, N. (2023). Effect of pre-and post-harvest factors on 'Benny' Valencia fruit rind phenolics on mitigation of chilling and non-chilling disorders during cold storage. *Journal of Horticulture and Postharvest Research*, 6(3), 299-316. https://doi.org/10.22077/jhpr.2023.6387.1319
- Baswal, A.K., Dhaliwal, H.S., Singh, Z., Mahajan, B.V.C., & Gill, K.S. (2020). Postharvest application of methyl jasmonate, 1-methylcyclopropene and salicylic acid extends the cold storage life and maintain the quality of 'Kinnow' mandarin (*Citrus nobilis* L. X *C. deliciosa* L.) fruit. *Postharvest Biology and Technology*, 161, 111064. https://doi.org/10.1016/j.postharvbio.2019.111064
- Fan, Z., Lin, B., Lin, Y., Chen, Yazhen., Chen, G., Chen, J., Wang, H., Chen, Yihui., & Lin, H. (2024). Alleviation of chilling injury in cold-stored Chinese olive (*Canarium album* Lour.) fruit by γ-aminobutyric acid treatment in relation to ROS metabolism. *Scientia Horticulturae*, 327, 112851. https://doi.org/10.1016/j.scienta.2024.112851
- Ghorbani, B., Pakkish, Z., & Khezri, M. (2018). Nitric oxide increases antioxidant enzyme activity and reduces chilling injury in orange fruit during storage. *New Zealand Journal of Crop and Horticultural Science*, 46(2): 101-116. https://doi.org/10.1080/01140671.2017.1345764
- Habibi, F., Ramezanian, A., Guillén, F., Serrano, M., & Valero, D. (2020). Blood oranges maintain bioactive compounds and nutritional quality by postharvest treatments with γ-aminobutyric acid, methyl jasmonate or methyl salicylate during cold storage. *Food Chemistry*, *306*, 125634. https://doi.org/10.1016/j.foodchem.2019.125634



- Habibi, F., Ramezanian, A., Rahemi, M., Eshghi, S., Guillén, F., Serrano, M., & Valero, D. (2019). Postharvest treatments with γ-aminobutyric acid, methyl jasmonate, or methyl salicylate enhance chilling tolerance of blood orange fruit at prolonged cold storage. *Journal of the Science of Food and Agriculture*, 99, 6408–6417. https://doi.org/10.1002/jsfa.9920
- Huang, R.H., Liu, J.H., Lu, Y.M., & Xia, R.X. (2008). Effect of salicylic acid on the antioxidant system in the pulp of 'Cara Cara' navel orange (*Citrus sinensis* L. Osbeck) at different storage temperatures. *Postharvest Biology and Technology*, 47, 168–175. https://doi.org/10.1016/j.postharvbio.2007.06.018
- Hui, G., Zheng, K.Z., Hong, K.C., Ni, C., Yue, Y., Dan, N.W., & Wei, C. (2016). Melatonin treatment delays postharvest senescence and regulates reactive oxygen species metabolism in peach fruit. *Postharvest Biology and Technology*, 118, 103–110. https://doi.org/10.1016/j.postharvbio.2016.03.006
- Jannatizadeh, A. (2019). Exogenous melatonin applying confers chilling tolerance in pomegranate fruit during cold storage. *Scientia Horticulturae*, 246, 544–549. https://doi.org/10.1016/j.scienta.2018.11.027
- Jimenez, A., Creissen, G., Kular, B., Firmin J., Robinson, S., Verhoeyen, M., & Mullineaux, P. (2002). Changes in oxidative processes and components of the antioxidant system during tomato fruit ripening. *Planta*, 214, 751–758.
- Khorshidi, Sh., Davarynejad, Gh., Tehranifar, A., & Fallahi, E. (2011). Effect of modified atmosphere packaging on chemical composition, antioxidant activity, anthocyanin, and total phenolic content of cherry fruits. *Horticulture and Environment Biotechnology*, *52*, 471-481.
- Lado, J., Gambetta, G., & Zacarias, L. (2018). Key determinants of citrus fruit quality: Metabolites and main changes during maturation. *Scientia Horticulturae*, 233, 238-248. https://doi.org/10.1016/j.scienta.2018.01.055
- Lara, I., Belge, B., & Goulao, L.F. (2014). The fruit cuticle as a modulator of postharvest quality. *Postharvest Biology and Technology*, 87, 103–112. https://doi.org/10.1016/j.postharvbio.2013.08.012
- Liao, L., Li, J., Lan, X., Li, Yaman, Li, Yunjie, Huang, Z., Jin, Z., Yang, Y., Wang, X., Zhang, M., Sun, G., Zhang, X., Xiong, B., & Wang, Z. (2024). Exogenous melatonin and interstock treatments confer chilling tolerance in citrus fruit during cold storage. *Scientia Horticulturae*, 327, 112802. https://doi.org/https://doi.org/10.1016/j.scienta.2023.112802
- Liu, S., Huang, H., Huber, D.J., Pan, Y., Shi, X., & Zhang, Z. (2020). Delay of ripening and softening in 'Guifei'mango fruit by postharvest application of melatonin. *Postharvest Biology and Technology*, 163, 111136. https://doi.org/10.1016/j.postharvbio.2020.111136
- Lorente-Mento, J. M., Guillén, F., Martínez-Romero, D., Carrión-Antoli, A., Valero, D., & Serrano, M. (2023). γ-Aminobutyric acid treatments of pomegranate trees increase crop yield and fruit quality at harvest. *Scientia Horticulturae*, 309, 111633. https://doi.org/10.1016/j.scienta.2022.111633
- Ma, Q., Lin, X., Wei, Q., Yang, X., Zhang, Y., & 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
- Makule, E., Dimoso, N., & Tassou, S.A. (2022). Precooling and cold storage methods for fruits and vegetables in Sub-Saharan Africa: A Review. *Horticulturae*, 8(9), 776. https://doi.org/10.3390/horticulturae8090776
- Mukhim, C., Nath, A., Deka, B.C., & Swer, T.L. (2015). Changes in physico-chemical properties of Assam lemon (*Citrus limon* Burm.) at different stages of fruit growth and development. *Bioscan*, 10(2), 535–537.
- Ngaffo Mekontso, F., Duan, W., Cisse, E.H.M., Chen, T., & Xu, X. (2021). Alleviation of postharvest chilling injury of carambola fruit by γ-aminobutyric acid: Physiological, biochemical, and structural characterization. *Frontiers in Nutrition*, *891*. https://doi.org/10.3389/fnut.2021.752583
- Ramesh, S.A., Tyerman, S.D., Xu, B., Bose, J., Kaur, S., Conn, V., Domingos, P., Ullah, S., Wege, S., & Shabala, S. (2015). GABA signalling modulates plant growth by directly regulating the activity of plant-specific anion transporters. *Nature communications*, *6*, 78-79.

- Rapisarda, P., Bellomo, S.E., & Intelisano, S. (2001). Storage temperature effects on blood orange fruit quality. *Journal of Agricultural and Food Chemistry*, 49, 3230–3235. https://doi.org/10.1021/jf0100321
- Rasouli, M., Saba, M.K., & Ramezanian, A. (2019). Inhibitory effect of salicylic acid and Aloe vera gel edible coating on microbial load and chilling injury of orange fruit. *Scientia Horticulturae*, 247, 27-34. https://doi.org/10.1016/j.scienta.2018.12.004
- 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
- Rey, F., Zacarías, L., & Rodrigo, M.J. (2020). Carotenoids, vitamin C, and antioxidant capacity in the peel of mandarin fruit in relation to the susceptibility to chilling injury during postharvest cold Storage. *Antioxidants*, 9(12), 1296. https://doi.org/10.3390/antiox9121296
- Sayyari, M., Babalar, M., Kalantari, S., Martínez-Romero, D., Guillén, F., Serrano, M., & Valero, D. (2011). Vapour treatments with methyl salicylate or methyl jasmonate alleviated chilling injury and enhanced antioxidant potential during postharvest storage of pomegranates. *Food Chemistry*, 124, 964–970. https://doi.org/10.1016/j.foodchem.2010.07.036
- Serna-Escolano, V., Martínez-Romero, D., Giménez, M. J., Serrano, M., García-Martínez, S., Valero, D., & Zapata, P. J. (2021). Enhancing antioxidant systems by preharvest treatments with methyl jasmonate and salicylic acid leads to maintain lemon quality during cold storage. *Food Chemistry*, 338, 128044. https://doi.org/10.1016/j.foodchem.2020.128044
- Siboza, X.I., Bertling, I., & Odindo, A.O. (2014). Salicylic acid and methyl jasmonate improve chilling tolerance in cold-stored lemon fruit (Citrus limon). *Journal of Plant Physiology*, *171*, 1722–1731. https://doi.org/10.1016/j.jplph.2014.05.012
- Sun, Y., Singh, Z., Tokala, V.Y., & Heather, B. (2019). Harvest maturity stage and cold storage period influence lemon fruit quality. *Scientia Horticulturae*, 249, 322–328. https://doi.org/10.1016/j.scienta.2019.01.056
- Tarancón, P., Tárrega, A., González, M., & Besada, C. (2021). External quality of mandarins: influence of fruit appearance characteristics on consumer choice. *Foods*, 10(9), 2188. https://doi.org/10.3390/foods10092188
- Vidal, A., Talens, P., Prats-Montalba'n, J.M., Cubero, S., Albert, F., & Blasco, J. (2013). In-line estimation of the standard colour index of citrus fruits using a computer vision system developed for a mobile platform. *Food Bioprocess Technology*, *6*, 3412–3419.
- Wang, S.-Y., Shi, X.-C., Liu, F.-Q., & Laborda, P. (2021). Effects of exogenous methyl jasmonate on quality and preservation of postharvest fruits: A review. *Food Chemistry*, 353, 129482. https://doi.org/10.1016/j.foodchem.2021.129482
- Wu, C., Hao, W., Yan, L., Zhang, H., Zhang, J., Liu, C., & Zheng, L. (2023). Postharvest melatonin treatment enhanced antioxidant activity and promoted GABA biosynthesis in yellow-flesh peach. *Food Chemistry*, 419, 136088. https://doi.org/10.1016/j.foodchem.2023.136088
- Yılmaz, M. (2024). Enhancing cold storage quality of blueberry (*Vaccinium corymbosum* 'Bluecrop') with methyl jasmonate treatments. *Applied Fruit Science*, 66, 81–88. https://doi.org/10.1007/s10341-023-01020-5
- Ze, Y., Gao, H., Li, T., Yang, B., & Jiang, Y. (2021). Insights into the roles of melatonin in maintaining quality and extending shelf life of postharvest fruits. *Trends in Food Science & Technology*, 109, 569–578. https://doi.org/10.1016/j.tifs.2021.01.051
- Zhang, Y., Tang, H., Lei, D., Zhao, B., Zhou, X., Yao, W., & Zhang, Y. (2023). Exogenous melatonin maintains postharvest quality in kiwiberry fruit by regulating sugar metabolism during cold storage. *LWT*, 174, 114385. https://doi.org/https://doi.org/10.1016/j.lwt.2022.114385

