



Effect of Melatonin on Alleviating Chilling Injury in Cold-Stored 'Kensington Pride' Mangoes: A Comparative Evaluation of Long vs Short Dip Time

Mekhala Dinushi Kananke Vithana¹ · Hafiz Muhammad Shoaib Shah¹ · Mahmood Ul Hasan¹ · Zora Singh¹

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Abstract

Mangoes (*Mangifera indica* L.) are notably susceptible to chilling injury (CI) when stored below 13 °C. This temperature threshold is too high for extended storage. Therefore, the influence of postharvest immersion in melatonin (MT) (50, 75, 100 $\mu\text{mol L}^{-1}$) for 120 min on CI in 'Kensington Pride' mangoes kept at 5 ± 1 °C for 21 or 28 d was investigated. The most effective MT concentration was 75 $\mu\text{mol L}^{-1}$ causing 36.3% reduction in CI incidence compared to control. To facilitate adoptability in commercial practice, the influence of 120 min vs. 5- and 3-min immersions in 75 $\mu\text{mol L}^{-1}$ MT was also tested. The highest efficacy in reducing CI was shown by mangoes immersed in 75 $\mu\text{mol L}^{-1}$ MT for 5 min with 52.2% and 38.6% lower CI incidence and CI index, respectively, than the control. This improved chilling tolerance in MT-treated mangoes was achieved through reduced hydrogen peroxide (H_2O_2) radicals, malondialdehyde (MDA) production, relative electrolyte leakage (REL) and lipoxygenase (LOX) activity while higher ascorbic acid (AsA) and glutathione (GSH) levels, and activities of ascorbate peroxidase (APX), glutathione reductase (GR) and monodehydroascorbate reductase (MDHAR). Fruit colour, soluble solids content (SSC), and titratable acidity (TA) were maintained in 75 $\mu\text{mol L}^{-1}$ MT treated fruit. MT was effective in reducing CI in mangoes up to 21 d at 5 ± 1 °C. Overall, immersing 'Kensington Pride' mangoes in 75 $\mu\text{mol L}^{-1}$ MT for 5 min before cold storage (5 ± 1 °C) shows high adoption potential to mitigate CI.

Keywords Chilling tolerance · Redox homeostasis · *Mangifera indica* L. · Plant growth regulators · Postharvest · Ascorbate-glutathione cycle

Introduction

Mango (*Mangifera indica* L.) is rich in anticarcinogenic metabolites such as lupeol, mangiferin and phenolic acids (Vithana et al. 2018a). The levels of lupeol and other anticarcinogenic phenolic compounds can be significantly increased if mangoes are stored at or below 5 °C due to cold-induced elicitation of their biosynthesis (Vithana et al. 2018a). Besides, in modern health-conscious markets, there

is a high demand, and guaranteed attractive prices for fruit rich in health-promoting compounds. Despite these advantages, mangoes cannot be stored at 5 °C for a longer period due to postharvest chilling injury (CI) incidence (Brecht 2019). If mangoes harvested at commercial maturity are stored below 13 °C than 2 weeks, they suffer CI, which makes them unsaleable due to skin browning, darkened lentils, greying, pitting and developing sunken areas on the fruit skin (Vithana et al. 2025).

Improving chilling tolerance of mango fruit has been a foremost research challenge in the recent past, which has been compiled by Sati et al. (2025). The mitigation of CI facilitates accessibility to long-distance markets and extended marketing windows of fresh mango fruit. The steadfast movement of the global horticulture industry towards net-zero emissions and sustainable production technologies has led scientists to discover more environmentally friendly approaches to extend the storage life of

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✉ Mekhala Dinushi Kananke Vithana
m.vithana@ecu.edu.au

¹ Horticulture and Centre for Crops and Horticulture Analytics for Nutrition and Climate Excellence, School of Science, Edith Cowan University, 270, Joondalup Drive, Joondalup, WA, Australia

fresh produce. In line with this trend, the application of melatonin (MT), food safe and eco-friendly phytohormone, has been explored to evaluate its efficacy in modulating fruit quality, ripening and storage life of some mango cultivars (Liu et al. 2020; Rastegar et al. 2020; Njie et al. 2022, 2023; Bhardwaj et al. 2021, 2022a, b; Xu et al. 2023; Kebbeh et al. 2023). MT is known to reduce CI through different modes of actions. Analysing several previous studies, Shah et al. (2024) have summarised that exogenous MT application before cold storage may induce an earlier accumulation of H_2O_2 , triggering antioxidant enzymes and phenolic biosynthesis in the treated tissues. This initial increase in hydrogen peroxide (H_2O_2) results in higher biosynthesis of endogenous MT. It subsequently triggers the ascorbate-glutathione (AsA-GSH) pathway (Fig. 4). Increased activity of AsA-GSH cycle critically contributes to the detoxification of H_2O_2 at the latter stage of cold storage. Additionally, exogenous MT application results in higher proline content through enhanced activities of Δ^1 -pyrroline-5-carboxylate synthetase and ornithine- δ -aminotransferase and reduced activity of proline dehydrogenase (Bhardwaj et al. 2021). Higher accumulation of endogenous polyamines and γ -aminobutyric acid (GABA) and higher GABA shunt activity (Glutamate decarboxylase, GABA-transaminase) also contribute to MT induced chilling tolerance in mango (Bhardwaj et al. 2022b). Accordingly, the positive impact of MT in alleviating CI in cold-stored ($4-5^\circ\text{C}$) mangoes has been reported on different cultivars: ‘Dashehari’, ‘Langra’, ‘Chaunsa’ (Bhardwaj et al. 2022a, b), ‘Guifei’ (Xu et al. 2023) and ‘Keitt’ (Kebbeh et al. 2023). These studies have reported a range of concentrations of MT for alleviating CI in different mango cultivars. The optimum concentration of MT for the effective CI control in ‘Dashehari’, ‘Langra’, ‘Chaunsa’ (Bhardwaj et al. 2022b) and ‘Keitt’ mangoes (Kebbeh et al. 2023) was reported as $100\ \mu\text{mol L}^{-1}$. A concentration of $500\ \mu\text{mol L}^{-1}$ worked best for ‘Guifei’ mangoes (Xu et al. 2023). Additionally, the optimum submerge duration reported for ‘Dashehari’, ‘Langra’ and ‘Chaunsa’ mangoes was 120 min (Bhardwaj et al. 2022a, b). ‘Guifei’ and ‘Keitt’ mangoes performed better when immersed in MT solutions for 60 and 30 min, respectively (Xu et al. 2023; Kebbeh et al. 2023). However, cultivar ‘Gulab Jamun’ was found to be non-responsive to MT application for any duration (Bhardwaj et al. 2022b). As the same studies reported that CI was significantly reduced in cultivar ‘Langra’ up to 28 d (Bhardwaj et al. 2022b) and in cultivar ‘Guifei’ (Xu et al. 2023) up to 40 d, a research need was prompted to investigate the responsiveness of major Australian mango cv. ‘Kensington Pride’ to the application of MT in reducing CI. Besides, the reported studies have used 30 to 120 min postharvest immersions. This lengthy submergence may impose practical challenges due to possible interferences with general

postharvest practices at a packing house (Brecht et al. 2020). Additionally, this may induce unwanted physiological processes such as anaerobic respiration, adversely impacting fruit quality and storage life in susceptible mango cultivars.

Accordingly, this study aimed to investigate the responsiveness of mango cultivar ‘Kensington Pride’ to MT submersion and to explore the efficacy of MT in reducing CI at variable short and long dipping times to improve its practicality. ‘Kensington Pride’ was selected as the preferred cultivar due to its abundance (43%) in Australian mango industry and higher susceptibility to CI.

Materials and Methods

Two independent experiments were carried out to investigate the effects of MT in reducing CI in cold-stored ‘Kensington Pride’ mango fruit and to explore the possibility of shortening the fruit submersion time in MT to examine its commercial adoption potential. The first experiment was designed to identify the optimum MT concentration to reduce CI when submerged for 120 min (Bhardwaj et al. 2022a, b). The second experiment was conducted to compare the efficacy of short duration immersions that may possibly facilitate its use in conjunction with current postharvest practices (5 and 3 min) as compared to the previously reported lengthy immersion (120 min), tested under experiment 1.

Experiment 1: Effects of Different Concentrations of MT ($50, 75$ and $100\ \mu\text{mol L}^{-1}$) on CI in ‘Kensington Pride’ Mango Fruit Cold-stored ($5 \pm 1^\circ\text{C}$) for 21 and 28 d

Source of mangoes and experimental plan:

‘Kensington Pride’ mangoes at commercial harvest maturity {hard mature green, light cream colour pulp, average firmness: 153.8 N; average soluble solids content (SSC): 10.7%} were manually picked from a commercial orchard at Gingin, Western Australia ($31^\circ 27' \text{S}$, $115^\circ 55' \text{E}$) during February 2023. Similar sized fruit with a good appearance were selected and packed as single layers in cardboard boxes with lids. These boxes were subsequently brought to the Horticulture Laboratory at the School of Science, Edith Cowan University, Joondalup, Western Australia, 6027 in less than 2 h under a set temperature and relative humidity (RH) of 15°C and 60% respectively. The experiment was laid out using a Completely Randomised Design (CRD) with a two-factor factorial arrangement (MT concentrations and cold storage durations). The treatments and control had four replications each with 16 mangoes per replication. Before arbitrarily separating into treatments and the control, all the mangoes were thoroughly washed with tap water

to remove any adhered dust or dirt. Then the experimental mangoes, split into different treatment lots, were submerged for 120 min either in 50, 75 or 100 $\mu\text{mol L}^{-1}$ MT solutions at room temperature (21 ± 1 °C). Required amount of MT was first dissolved in pure ethanol at a rate of 50 mg MT per 1 mL of ethanol according to the manufacturer's instructions. The mixing was done in an amber glass bottle. The bottle was shaken until a completely clear solution was obtained. Then, this MT solution was added to the required volume of water at neutral pH to prepare the above concentrations. The aqueous solution preparation and fruit submersion were carried out under subdued light conditions to avoid photo-degradation of MT. Control mangoes were not subjected to any aqueous dip treatment or light control other than initial thorough washing with tap water. Upon completion of the soaking in MT solutions at different concentrations, treated and control mangoes were air dried, packed into open top cardboard boxes and stored in the dark at 5 ± 1 °C and $80 \pm 5\%$ RH for either 21 or 28 d. After the respective storage period (21 or 28 d), the fruit were removed from cold storage and placed in the laboratory (21 ± 1 °C) for 6 d. CI index and CI incidence were recorded on the day of removal from cold storage (21 or 28 d) and 3 d and 6 d after each storage period. The fruit colour, SSC, titratable acidity (TA), relative electrolyte leakage (REL) and AsA were estimated using fresh samples upon completion of cold storage and shelf periods ($21 + 6$ d and $28 + 6$ d). The oxidative and anti-oxidative parameters including hydrogen peroxide (H_2O_2), malondialdehyde (MDA), superoxide dismutase (SOD), peroxidase (POD), catalase (CAT), lipoxygenase (LOX), polyphenol oxidase (PPO), phenylalanine ammonia-lyase (PAL), total phenolic content (TPC), AsA, GSH, glutathione reductase (GR), ascorbate peroxidase (APX), monodehydroascorbate reductase (MDHAR) and dehydroascorbate reductase (DHAR) were assessed using cryopreserved representative samples stored at -80 ± 1 °C.

Experiment 2: Effects of Different Submerge Durations (120, 5 and 3 min) in 75 $\mu\text{mol L}^{-1}$ MT on CI in 'Kensington Pride' Mango Fruit Cold-stored (5 ± 1 °C) for 21 d

Source of mangoes and experimental plan:

'Kensington Pride' mangoes at harvest maturity, as explained under experiment 1, were manually picked from the same orchard and brought to the Horticulture Laboratory under the same conditions as detailed under experiment 1 (2.1). Experiment 2 followed a Completely Randomised Design (CRD) with one factor (submerge duration). Mangoes were washed with tap water and split into treatment units and control (4 replications, 16 mangoes per replication) as described under 2.1. Then the mangoes belonging

to different treatment units were submerged in 75 $\mu\text{mol L}^{-1}$ MT solution (optimum concentration revealed in experiment 1) at room temperature (21 ± 1 °C) for either 120, 5 or 3 min. Similar to experiment 1, the control fruit were not subjected to any aqueous dip treatment or light control other than initial washing. Cold storage conditions and all the parameters analysed were the same as with experiment 1 (2.1). However, in experiment 2, mango fruit were stored only for 21 d at 5 ± 1 °C and $80 \pm 5\%$ relative humidity (RH) before subsequent shelf period of 6 d at ambient temperature as MT treatment was not able to control CI incidence or reduce CI index in experiment 1 when stored for 28 d, both treated and control mangoes recording over 90% CI incidence and statistically non-significant difference ($P \leq 0.05$) in CI index.

Chemicals

Melatonin (98% pure) was bought from Merck Pty Ltd, Mountain Highway, Bayswater, Victoria (3153), Australia, while all the reagents and other chemicals were purchased from ROWE Scientific Pty Ltd, Wangara (6065), Western Australia.

Determination of CI Parameters

CI incidence (%) and CI index in eight fruit per replication were recorded using common CI symptoms of mangoes such as noticeable lenticels and greyish areas on the peel using the rating scale; 0- if no visual symptoms of CI, 1- very low occurrence of CI symptoms ($< 5\%$), 2- light CI ($5\text{--}10\%$), 3- moderate CI ($10\text{--}25\%$) and 4- severe CI ($> 25\%$), and calculated using the same methods reported by Vithana et al. (2025).

Analysis of Quality Parameters

Colour

The colour coordinates (a^* , b^* and L^*) of the opposite faces of mango fruit upon completion of 6 d on shelf were recorded in 8 fruit per replication using ColorFlex EC HunterLab colour spectrophotometer (Hunter Associates Laboratory Inc, 11491 Sunset Hills Road, Reston, Virginia, 20190, USA). Then the hue angle (h°) and chromaticity (chroma) were worked out as below to determine the full colour profile (Vithana et al. 2024).

$$\text{Hue angle}(h^\circ) = \arctan \left(b^*/a^* \right)$$

$$\text{Chroma} = \sqrt{a^{*2} + b^{*2}}$$

SSC, TA and SSC and TA Ratio

To determine SSC (%) the prism of the refractometer (Atago-Palette PR 101, Atago CO. Ltd, Itabashi-Ku, and Tokyo, Japan) was filled with a fresh composite mango juice sample extracted from 8 fruit per replication and passed through a muslin cloth. TA (%) of the same juice samples was determined using the same method described by Sakimin (2011). Then the SSC: TA ratio was calculated as: SSC: TA=SSC (%) / corresponding TA (%).

Weight Loss (%)

The percentage weight loss was calculated for the same fruit that were used to calculate CI incidence and CI index in both experiments.

$$\text{Weight loss (\%)} = \frac{(\text{Initial weight} - \text{Weight at date of measurement})}{(\text{Initial weight})} \times 100$$

Determination of Oxidative Stress and Membrane Damage

To determine the level of H_2O_2 , the method described by Shah et al. (2023) was used. In brief, representative pulp or peel samples (1 g) from 8 fruit (at 6 d shelf period) from each replication was extracted with 1 mL of 1% trichloroacetic acid (TCA) and centrifuged (4 °C, 12,000 $\times g$, 15 min). Next, 100 μL of supernatant, 100 μL of 10 mmol L^{-1} phosphate buffer (pH 7.0) and 1000 μL of 1 M potassium iodide were mixed and the absorbance at 390 nm was obtained (UV–VIS, Jenway, Model 7205, UK) and presented as $\mu\text{mol kg}^{-1}$.

To determine the level of MDA, the same method was used as detailed by Yao et al. (2009). Firstly, a representative composite pulp/ peel sample (1 g) was extracted with 10% (w/v) cold TCA solution (10 mL) prior to centrifugation (10,000 $\times g$, 20 min). The supernatant was heated in a hot water bath at 90 °C (15 min) before being centrifuged (10,000 $\times g$, 20 min) with 2 mL of 2-thiobarbituric (TBA) solution (0.6% w/v). The absorbance of the supernatant was noted at 532 nm (for MDA) and at 600 nm for MDA-TBA adduct (UV–visible spectrophotometer- Jenway, model 7205, UK). Finally, the MDA concentration was quantified as nmol kg^{-1} .

REL was determined by the method reported earlier by Hasan et al. (2025) using representative peel samples from 8 fruit per replicate. Firstly, 20 peel Sections (10 $\pm$ 1 mm diameter) were put into 30 mL deionised water and kept at 25 °C for 30 min. After 30 min, the initial electrical conductivity reading was recorded (Digital conductivity meter, HI-98304, Hanna, Mauritius). Then the samples were boiled

in a water bath for 15 min, and the final reading was taken after cooling. REL percentage was calculated as:

$$\text{REL \%} = \frac{\text{Initial electrical conductivity}}{\text{Final electrical conductivity}} \times 100$$

Enzyme Assays

The activities of antioxidant enzymes SOD, POD and CAT were determined using the methods earlier described by Shah et al. (2017), whilst PAL activity was determined by the method used by Khan et al. (2003). The protocol used by Chen et al. (2022) was used to assess the activity of enzyme LOX and the PPO activity. For enzyme assays (SOD, POD, CAT, LOX, PPO), a crude extract was prepared using a composite sample of mango pulp/ peel (from 8 fruit from each replication that completed 6 d shelf period) homogenised with potassium citrate buffer (cold, pH 4.0, 4 mL) followed by centrifugation (10,000 $\times g$ for 20 min, at 4 °C). Then the supernatant of this crude enzyme extract was used for the determination of enzymes mentioned above. For the assessment of PAL activity, a different crude enzyme extract was prepared by mixing 1 g of a composite mango pulp/ peel sample homogenised in Tris-HCL buffer (ice cold, 50 mmol, pH 8.5 with 5% w/v polyvinylpyrrolidone and β -mercaptoethanol, 14.4 mmol L^{-1}). The activities of all these enzymes were determined on a protein basis (Bradford, 1976) by recording absorbance using a UV–VIS spectrophotometer- Jenway, model 7205, UK, and presented as $\text{mmol kg}^{-1} \text{ s}^{-1}$.

AsA-GSH Cycle

AsA

Determined using the same method reported by Malik and Singh (2005) and reported as mg kg^{-1} (fresh weight basis).

GSH

Determined with the method detailed by Ge et al. (2015). In summary, a neutralized (pH 7.0) supernatant (200 μL) of homogenised {10 mL TCA solution (5% w/v)} and centrifuged (12,000 $\times g$, 4 °C, for 15 min) composite mango pulp sample (1 g) was reacted with a mixture of phosphate buffer {0.1 mol, pH 7.5, 1100 μL , with EDTA (5 mmol)}, nicotinamide adenine dinucleotide phosphate (NADPH 1 mmol, 200 μL) and 5,5'-dithiobis-(2-nitrobenzoic acid, 6 mmol, 100 μL). After 5 min, the absorbance at 412 nm was measured and the total glutathione content was reported as mg kg^{-1} (fresh weight basis).

GR, MDHAR, DHAR and APX

The activities of enzymes GR, MDHAR and DHAR were determined as detailed by Chumyam et al. (2017), whereas the APX activity was assessed following the protocol used by Shah et al. (2017). For all enzyme assays (GR, MDHAR, DHAR, APX), a crude enzyme extract was made by homogenising and centrifuging (25 min at 15,000 $\times g$, 4 °C) 1 g of composite mango pulp/ peel sample (from 8 fruit per replication, upon completion of the 6 d shelf period) with a reaction mixture (10 mL) {potassium phosphate buffer (100 mmol, pH 7.0), polyvinyl pyrrolidone (PVP, 2.5%), EDTA (1 mmol) and L-ascorbate (1 mmol)}. Then the supernatant was used in all enzyme assays. The absorbance value was measured with UV–visible spectrophotometer (Jenway, model 7205, UK) and used for calculations of enzyme activities. The activities of all these enzymes were determined on a protein basis (Bradford, 1976) by recording absorbance using a UV–VIS spectrophotometer- Jenway, model 7205, UK, and presented as $\text{mmol kg}^{-1} \text{ s}^{-1}$.

Determination of Total Phenolic content (TPC)

TPC was determined using Folin-Ciocalteu reagent adopting the same method described by Vithana et al. (2018b) and presented as g kg^{-1} (fresh weight basis).

Statistical Analysis

For the experiment 1, two-way ANOVA was used to determine the statistical significance of MT treatments, cold storage durations (21 and 28 d) and their interactions (MT $\times$ cold storage duration), while one-way ANOVA was used to analyse the statistical significance of submerge duration in experiment 2. Duncan's Multiple Range Test ($P \leq 0.05$) was used in both experiments to compare means. Genstat statistical system (24th edition, VSN International, Hemel Hempstead, UK) was used in both experiments. In experiment 1 results, upper case letters show the statistical difference among the storage durations and lowercase letters indicate the statistical significance among MT treatments and control. In experiment 2 results, lowercase letters indicate the statistical significance among MT submerge durations and control. The results were conveyed as means $\pm$ standard deviation (SD) in both experiments.

Results

Experiment 1: Effects of Different Concentrations of MT (50, 75 and 100 $\mu\text{mol L}^{-1}$) on CI in 'Kensington Pride' Mango Fruit Cold-stored (5 ± 1 °C) for 21 and 28 d

Effects of Different Concentrations of MT on CI Incidence and CI Index

The influence of MT on CI incidence and CI index varied along the shelf period (Table 1). By the end of the total shelf period of 6 d, mangoes immersed in 75 $\mu\text{mol L}^{-1}$ MT and stored for 21 d recorded significant reductions in CI incidence (36.3%) and CI index (29.3%) compared to the untreated control (Table 1). MT was not effective in reducing either CI incidence or CI index in mangoes stored for 28 d (Table 1). Thus, there was a significant effect of storage duration on CI incidence, having a 34.2% higher occurrence in the mangoes stored for 28 d compared to 21 d (Table 1).

Effects of Different Concentrations of MT on the Quality Parameters (weight loss, colour, SSC, TA and SSC: TA ratio)

The mangoes stored for 28 d recorded a significantly higher weight loss (43.2%) compared to mangoes stored for 21 d at the end of the shelf period (Table 1). There was a significant effect of cold storage duration on all colour parameters recording 58.1% higher redness (a^*), 17.5% lower yellowness (b^*), 25.0% lower lightness (L^*), 17.8% lower colour saturation (chroma) and 3% lower hue° in mango fruit stored for 28 d compared to the fruit stored for 21 d (Table 2). After the shelf period (6 d), mangoes submerged in 50 and 100 $\mu\text{mol L}^{-1}$ MT and cold-stored for 21 d showed 21.0% and 21.6% lower SSC values (Table 2). Fruit immersed in 100 $\mu\text{mol L}^{-1}$ MT recorded a significantly higher SSC (%) compared to control when stored for 28 d (Table 2). Additionally, there was a significant interaction between MT concentration and cold storage duration (Table 2). TA was 58.3% less in fruit cold stored for 28 d compared to 21 d (Table 2). Due to lower SSC (%) values mentioned above, SSC: TA ratio was 35.9% and 39.3% lower in the fruit submerged in 50 and 100 $\mu\text{mol L}^{-1}$ MT respectively compared to untreated mangoes when stored for 21 d (Table 2). Due to significantly higher acidity, the SSC: TA ratio was significantly lower (55.8%) in the fruit cold-stored for 21 d compared to 28 d (Table 2).

Table 1 Effects of different concentrations of MT on CI incidence, CI index and weight loss in 'Kensington Pride' mangoes cold-stored ($5 \pm 1^\circ\text{C}$) for 21 or 28 d followed by an ambient shelf period of 6 d

Parameter	Cold storage and shelf period (d)									
CI incidence (%)	MT ($\mu\text{mol L}^{-1}$)	21+0d	28+0d	(Mean MT)	21+3d	28+3d	(Mean MT)	21+6d	28+6d	(Mean MT)
	0	31.2 $\pm$ 6.3	56.2 $\pm$ 14.0	43.7b	46.9 $\pm$ 5.4	75.0 $\pm$ 8.8	60.9b	68.8 $\pm$ 6.2	96.9 $\pm$ 5.4	82.8b
	50	18.7 $\pm$ 14.0	55.2 $\pm$ 12.6	37.0ab	46.9 $\pm$ 10.4	68.8 $\pm$ 10.8	57.8ab	59.4 $\pm$ 5.4	93.8 $\pm$ 6.2	76.6b
	75	12.5 $\pm$ 12.5	43.8 $\pm$ 10.8	28.1a	34.4 $\pm$ 5.4	62.5 $\pm$ 8.8	48.4a	43.8 $\pm$ 10.8	90.6 $\pm$ 5.4	67.2a
	100	21.9 $\pm$ 13.6	60.8 $\pm$ 3.0	41.3ab	43.8 $\pm$ 14.0	71.9 $\pm$ 5.4	57.8ab	75.0 $\pm$ 8.8	93.8 $\pm$ 6.2	84.4b
	Mean (SP)	21.1 A	54.0B		43.0 A	69.5B		61.7 A	93.8B	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	MT	SP	MT $\times$ SP	MT	SP	MT $\times$ SP
		14.62	10.34	NS	11.43	8.08	NS	8.81	6.23	12.46
CI Index	0	0.19 $\pm$ 0.04	0.23 $\pm$ 0.05	0.21b	0.25 $\pm$ 0.04	0.30 $\pm$ 0.06	0.28b	0.41 $\pm$ 0.07	0.42 $\pm$ 0.12	0.41a
	50	0.14 $\pm$ 0.09	0.19 $\pm$ 0.06	0.17ab	0.26 $\pm$ 0.09	0.34 $\pm$ 0.06	0.30b	0.39 $\pm$ 0.10	0.40 $\pm$ 0.07	0.39a
	75	0.07 $\pm$ 0.07	0.18 $\pm$ 0.06	0.12a	0.18 $\pm$ 0.03	0.26 $\pm$ 0.08	0.22a	0.29 $\pm$ 0.08	0.38 $\pm$ 0.08	0.33a
	100	0.13 $\pm$ 0.08	0.21 $\pm$ 0.03	0.17ab	0.27 $\pm$ 0.12	0.36 $\pm$ 0.05	0.31b	0.41 $\pm$ 0.09	0.41 $\pm$ 0.06	0.41a
	Mean (SP)	0.13 A	0.20B		0.24 A	0.32B		0.37 A	0.40 A	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	MT	SP	MT $\times$ SP	MT	SP	MT $\times$ SP
		0.08	0.056	NS	0.08	0.063	NS	NS	NS	NS
Weight loss (%)	0	1.55 $\pm$ 0.18	7.90 $\pm$ 1.76	4.72b	4.90 $\pm$ 0.21	9.90 $\pm$ 2.04	7.40a	8.55 $\pm$ 0.26	13.93 $\pm$ 1.30	11.24a
	50	0.55 $\pm$ 0.15	8.00 $\pm$ 0.53	4.27b	4.40 $\pm$ 1.70	10.88 $\pm$ 0.74	7.64a	7.75 $\pm$ 0.47	14.15 $\pm$ 0.79	10.95a
	75	0.47 $\pm$ 0.13	5.12 $\pm$ 1.64	2.80a	3.82 $\pm$ 0.40	8.82 $\pm$ 2.60	6.32a	7.00 $\pm$ 0.60	13.03 $\pm$ 1.8	10.01a
	100	0.80 $\pm$ 0.35	7.65 $\pm$ 0.48	4.22b	4.37 $\pm$ 0.26	10.50 $\pm$ 1.53	7.44a	7.28 $\pm$ 0.68	12.70 $\pm$ 1.41	9.99a
	Mean (SP)	0.84 A	7.17B		4.38 A	10.02B		7.64 A	13.45B	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	MT	SP	MT $\times$ SP	MT	SP	MT $\times$ SP
		0.73	1.04	NS	NS	1.23	NS	NS	0.82	NS

$N=4$ replicates with 16 fruit per replication. MT = melatonin concentration, SP = storage period, MT $\times$ SP = interaction. LSD = Least Significant Difference at ($P \leq 0.05$). Upper case letters show the statistical difference ($P \leq 0.05$) among the storage durations and lowercase letters indicate the statistical significance among MT treatments and control. Same letter denotes non-significance. The results were expressed as means $\pm$ standard deviation (SD)

Effects of Different Concentrations of MT on the Levels of H_2O_2 , MDA, REL, LOX and PPO

The levels of H_2O_2 in the peel were 45.0% and 20.1% (50 $\mu\text{mol L}^{-1}$), 52.6% and 42.7% (75 $\mu\text{mol L}^{-1}$) and 24.4% and 27.6% (100 $\mu\text{mol L}^{-1}$) reduced respectively when cold-stored for 21 or 28 d (Table 3). Among MT treatments, 75 $\mu\text{mol L}^{-1}$ showed the highest efficacy in reducing H_2O_2 in the peel. The pulp H_2O_2 concentrations were 30.6%, 39.7% and 44.2% lower when stored for 21 d, and 33.7%, 44.9% and 27.0% lower in the fruit stored for 28 d after submersion in 50, 75 and 100 $\mu\text{mol L}^{-1}$ respectively (Table 3). The concentration of H_2O_2 in the peel (66.1%) and pulp (47.2%) was significantly higher when the fruit were cold stored for 28 d compared to 21 d. There were 73.7%, 76.0% and 59.2% reductions in MDA levels in the peel when stored for 21 d, while 49.7%, 40.1% and 32.8% reductions were observed in the fruit stored for 28 d treated with 50, 75 and 100 $\mu\text{mol L}^{-1}$ respectively (Table 3). Similarly, the MDA levels in the pulp were 54.2%, 50.8% and 33.5% lower in the fruit stored for 21 d, while these levels were 48.8%, 33.8% and 19.2% lower in 28 d stored fruit previously submerged in 50, 75 and 100 $\mu\text{mol L}^{-1}$ MT respectively (Table 3). REL in the peel was significantly reduced by 75 $\mu\text{mol L}^{-1}$ MT

compared to control recording 43.8% (21 d) and 38.6% (28 d) reductions (Table 3). The submersions in MT (50, 75 and 100 $\mu\text{mol L}^{-1}$) were able to reduce LOX activity in the peel by 35.5%, 29.3% and 28.5% when stored for 21 d and by 31.9%, 36.5% and 53.5% when stored for 28 d respectively, (Table 3). Similarly, the LOX activity in the pulp was reduced by 26.7%, 35.0% and 39.2% in the fruit stored for 21 d and by 44.8%, 36.6% and 41.6% when stored for 28 d previously submerged in 50, 75 and 100 $\mu\text{mol L}^{-1}$ respectively (Table 3). PPO in the peel showed a 40.8% increase when stored for 28 d compared to 21 d (Table 3).

Effects of Different Concentrations of MT on the Performance of Antioxidant Enzymes (SOD, POD, PAL) and the Level of Total Phenolic Compounds (TPC)

All postharvest submersions in MT significantly increased the activity of enzyme SOD in the pulp of the mangoes stored for 21 d showing 15.8%, 30.4% and 5.9% higher activity respectively (Table 4). Only 75 $\mu\text{mol L}^{-1}$ (60.0%) and 100 $\mu\text{mol L}^{-1}$ (71.4%) treatments were able to increase SOD activity in the pulp of mangoes stored for 28 d (Table 4). The storage duration significantly affected the activity of SOD in both peel and pulp, recording 19.6% and 52.6%

Table 2 Effects of different concentrations of MT on quality parameters of ‘Kensington Pride’ mangoes cold-stored (5 ± 1 °C) for 21 or 28 d followed by an ambient shelf period of 6 d

Parameter	Cold storage and shelf period (d)			
Colour				
a*	MT	21+6	28+6	(Mean MT)
	($\mu\text{mol L}^{-1}$)			
	0	1.59 $\pm$ 1.54	3.0 $\pm$ 0.98	2.30a
	50	-0.29 $\pm$ 1.98	1.81 $\pm$ 1.38	0.76a
	75	2.28 $\pm$ 1.51	2.50 $\pm$ 1.15	2.39a
	100	0.22 $\pm$ 0.60	1.76 $\pm$ 0.78	0.99a
	Mean (SP)	0.95 A	2.27B	
b*	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
		NS	1.13	NS
	0	44.91 $\pm$ 0.90	36.31 $\pm$ 1.56	40.61a
	50	43.52 $\pm$ 0.73	36.13 $\pm$ 1.22	39.83a
	75	45.86 $\pm$ 1.09	37.45 $\pm$ 1.45	41.65a
	100	44.69 $\pm$ 2.72	36.93 $\pm$ 2.06	40.81a
	Mean (SP)	44.75B	36.93 A	
L*	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
		NS	1.39	NS
	0	62.38 $\pm$ 1.06	45.78 $\pm$ 2.75	54.08a
	50	60.59 $\pm$ 0.78	44.89 $\pm$ 1.59	52.74a
	75	63.0 $\pm$ 2.23	48.35 $\pm$ 1.58	55.68a
	100	62.16 $\pm$ 2.59	46.64 $\pm$ 2.57	54.40a
	Mean (SP)	62.16B	46.65 A	
Chroma	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
		NS	1.77	NS
	0	44.97 $\pm$ 0.94	36.45 $\pm$ 1.50	40.71a
	50	43.57 $\pm$ 0.70	36.20 $\pm$ 1.17	39.89a
	75	45.94 $\pm$ 1.12	37.55 $\pm$ 1.39	41.74aa
	100	44.70 $\pm$ 2.72	36.98 $\pm$ 2.07	40.84a
	Mean (SP)	44.79B	36.8 A	
Hue°	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
		NS	1.39	NS
	0	87.98 $\pm$ 1.94	85.22 $\pm$ 1.66	86.60a
	50	90.35 $\pm$ 2.63	87.07 $\pm$ 2.24	88.71a
	75	87.20 $\pm$ 1.86	86.15 $\pm$ 1.89	86.68a
	100	89.75 $\pm$ 0.80	87.28 $\pm$ 1.11	88.52a
	Mean (SP)	88.82B	86.43 A	
SSC (%)	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
		NS	1.6	NS
	0	19.75 $\pm$ 1.10	16.83 $\pm$ 1.39	18.29b
	50	15.62 $\pm$ 0.96	17.12 $\pm$ 2.39	16.38a
	75	19.35 $\pm$ 2.28	16.40 $\pm$ 0.76	17.88ab
	100	15.48 $\pm$ 0.40	18.73 $\pm$ 0.90	17.10ab
	Mean (SP)	17.55	17.27	
Acidity (%)	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
		1.44	NS	2.04
	0	0.68 $\pm$ 0.04	0.30 $\pm$ 0.07	0.49a
	50	0.99 $\pm$ 0.35	0.38 $\pm$ 0.04	0.68a
	75	0.79 $\pm$ 0.19	0.36 $\pm$ 0.03	0.58a
	100	0.90 $\pm$ 0.15	0.34 $\pm$ 0.04	0.62a
	Mean (SP)	0.84B	0.35 A	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
		NS	0.14	NS

Table 2 (continued)

Parameter	Cold storage and shelf period (d)			
Colour				
SSC: Acidity	0	29.0±2.46	58.0±10.51	43.5a
	50	18.6±7.94	45.8±9.71	32.2a
	75	25.5±5.54	45.7±5.35	35.6a
	100	17.6±3.19	55.9±8.41	36.8a
	Mean (SP)	22.7 A	51.4B	
	LSD ($P \leq 0.05$)	MT	SP	MT×SP
		NS	6.41	NS

$N = 4$ replicates with 16 fruit per replication. MT = melatonin concentration, SP = storage period, MT×SP = interaction. LSD = Least Significant Difference at ($P \leq 0.05$). Upper case letters show the statistical difference ($P \leq 0.05$) among the storage durations and lowercase letters indicate the statistical significance among MT treatments and control. Same letter denotes non-significance. The results were expressed as means $\pm$ standard deviation (SD)

higher activity in the fruit stored for 21 d when compared with 28 d (Table 4). The activity of enzyme POD in the pulp was significantly higher in the fruit submerged in 75 $\mu\text{mol L}^{-1}$ (31.4%) and 100 $\mu\text{mol L}^{-1}$ (30.6%) MT when stored for 28 d compared to the untreated group (Table 4). The activity of POD in the peel was significantly higher in the fruit submerged in 75 $\mu\text{mol L}^{-1}$ MT and in the control compared to 50 and 100 $\mu\text{mol L}^{-1}$ MT treatments after cold storage for 28 d (Table 4). The storage duration affected the activity of POD in both peel and pulp having significantly higher activity (33.4% and 28.0% respectively) in the fruit stored for 28 d when compared to 21 d (Table 4). The activity of enzyme PAL in the pulp was significantly higher in the fruit submerged in 50 $\mu\text{mol L}^{-1}$ (93.3%), 75 $\mu\text{mol L}^{-1}$ (90.0%) and 100 $\mu\text{mol L}^{-1}$ (90.0%) MT when cold stored for 28 d compared to control (Table 4). The TPC in the peel was significantly higher when submerged in 75 $\mu\text{mol L}^{-1}$ (16.0%, 36.4%) and 100 $\mu\text{mol L}^{-1}$ (7.9%, 14.3%) MT under both 21 d and 28 d storage durations respectively than untreated fruit (Table 4). The storage duration also significantly influenced the TPC in the peel recording a 28.6% higher value in the fruit stored for 21 d compared to 28 d (Table 4). All MT submersions significantly increased TPC in the pulp of the mangoes stored for 21 d showing 29.8%, 39.2% and 19.5% higher values compared to control (Table 4), whereas only 75 $\mu\text{mol L}^{-1}$ MT immersion was able to increase (44.0%) pulp TPC significantly when stored for 28 d (Table 4). Similarly, the TPC in the peel was significantly higher (22.6%) in the mangoes stored for 21 d compared to 28 d (Table 4).

Effects of Different Concentrations of MT on Ascorbate-glutathione (AsA-GSH) Cycle

The level of AsA in the peel was significantly higher in the fruit submerged in 75 $\mu\text{mol L}^{-1}$ (47.2%) and 100 $\mu\text{mol L}^{-1}$ (18.6%) MT when cold-stored for 21 d (Table 5). All three MT treatments increased AsA in the peel significantly (52.5%, 67.8% and 61.5% respectively) in the fruit stored

for 28 d compared to untreated control (Table 5). The level of AsA in the peel was 41.6% higher when stored for 21 d compared to 28 d (Table 5). Similarly, the content of AsA in the pulp was 53.7% higher when stored for 21 d compared to 28 d (Table 5). Former submersion in 50 and 75 $\mu\text{mol L}^{-1}$ MT significantly increased the GSH level in the peel by 23.5 and 15.5% when cold stored for 21 d (Table 5). All three MT concentrations (50, 75, 100 $\mu\text{mol L}^{-1}$) were significantly effective in increasing peel GSH level (60.3%, 79.4%, and 66.6% respectively) in the mangoes stored for 28 d (Table 5). Submersion in 50 and 75 $\mu\text{mol L}^{-1}$ MT significantly increased the GSH level by 43.3% and 52.1% in the pulp when cold stored for 28 d. All three MT submersions (50, 75, 100 $\mu\text{mol L}^{-1}$) were significantly effective in increasing pulp GSH level (18.2%, 38.3%, and 26.6% respectively) in the mangoes stored for 21 d (Table 5). The levels of GSH in both peel and pulp were significant affected showing 69.2% (peel) and 23.7% (pulp) higher values in the fruit stored for 21 d compared to 28 d (Table 5). The APX activity of the pulp was significantly increased by submersions in 50 (23.2%) and 75 $\mu\text{mol L}^{-1}$ MT (29.6%) when cold stored for 21 d (Table 5). All three MT submersions (50, 75, 100 $\mu\text{mol L}^{-1}$) increased the APX activity by 42.6%, 72.0% and 47.6% respectively compared to control when stored for 28 d (Table 5). The APX activity in both peel (40.7%) and pulp (48.8%) were higher in the fruit stored for 21 d compared to 28 d (Table 5). MT (50, 75, 100 $\mu\text{mol L}^{-1}$) submersions significantly increased the activity of enzyme GR in the peel when stored for 28 d recording 58.5%, 49.5% and 53.1% higher values respectively compared to untreated fruit (Table 5). The activity of GR in the pulp was significantly increased by MT submersions (50, 75, 100 $\mu\text{mol L}^{-1}$) under both cold storage durations (21 or 28 d) compared to control fruit (Table 5). This increase was 37.0%, 28.1% and 33.0% in the pulp of the fruit stored for 21 d and 36.9%, 54.0% and 41.6% when stored for 28 d respectively (Table 5). Additionally, the GR activity of both peel (26.3%) and pulp (32.4%) were significantly higher in

Table 3 Effects of different concentrations of MT on physiological markers of membrane integrity in ‘Kensington Pride’ mangoes cold-stored (5 ± 1 °C) for 21 or 28 d followed by an ambient shelf period of 6 d

Parameter	Peel				Pulp			
	Cold storage and shelf period (d)				Cold storage and shelf period (d)			
H_2O_2 ($\mu\text{mol kg}^{-1}$)	MT ($\mu\text{mol L}^{-1}$)	21+6	28+6	(Mean MT)	MT ($\mu\text{mol L}^{-1}$)	21+6	28+6	(Mean MT)
	0	1.96 $\pm$ 0.10	5.20 $\pm$ 0.14	3.58c	0	0.98 $\pm$ 0.12	1.80 $\pm$ 0.08	1.39c
	50	1.08 $\pm$ 0.15	4.15 $\pm$ 0.75	2.61b	50	0.68 $\pm$ 0.13	1.20 $\pm$ 0.12	0.94b
	75	0.93 $\pm$ 0.13	2.98 $\pm$ 0.23	1.95a	75	0.59 $\pm$ 0.12	0.99 $\pm$ 0.11	0.79a
	100	1.48 $\pm$ 0.05	3.76 $\pm$ 0.39	2.62b	100	0.55 $\pm$ 0.12	1.31 $\pm$ 0.24	0.93ab
	Mean (SP)	1.36 A	4.02B		Mean (SP)	69.8 A	1.32B	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
MDA (nmol kg^{-1})		0.40	0.28	0.56		0.14	0.05	0.1
	0	5.89 $\pm$ 0.8	12.85 $\pm$ 3.0	9.4b	0	7.75 $\pm$ 1.8	10.40 $\pm$ 1.9	9.1c
	50	1.55 $\pm$ 0.7	6.46 $\pm$ 2.4	4.0a	50	3.55 $\pm$ 0.4	5.33 $\pm$ 1.6	4.4a
	75	1.41 $\pm$ 0.4	7.70 $\pm$ 3.1	4.6a	75	3.81 $\pm$ 0.5	6.89 $\pm$ 0.2	5.4a
	100	2.40 $\pm$ 0.9	8.64 $\pm$ 1.9	5.5a	100	5.15 $\pm$ 0.3	8.40 $\pm$ 1.1	6.8b
	Mean (SP)	2.81 A	8.91B		Mean (SP)	5.06 A	7.76B	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
REL (%)		2.13	1.5	NS		1.4	1	NS
	0	37.7 $\pm$ 5.6	48.2 $\pm$ 15.2	43.0b	0	NA	NA	NA
	50	35.9 $\pm$ 7.2	42.7 $\pm$ 11.6	39.3b	50	NA	NA	NA
	75	21.2 $\pm$ 4.3	29.6 $\pm$ 12.3	25.4a	75	NA	NA	NA
	100	28.9 $\pm$ 4.5	37.1 $\pm$ 7.6	33.0ab	100	NA	NA	NA
	Mean (SP)	30.9	39.4		Mean (SP)	NA	NA	NA
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
LOX ($\text{mmol kg}^{-1} \text{ s}^{-1}$)		11.4	8.1	NS		NA	NA	NA
	0	7.37 $\pm$ 0.5	10.24 $\pm$ 1.1	8.80b	0	6.14 $\pm$ 0.7	8.5 $\pm$ 1.0	7.32b
	50	4.75 $\pm$ 0.9	6.97 $\pm$ 1.1	5.86a	50	4.50 $\pm$ 0.6	4.69 $\pm$ 0.7	4.60a
	75	5.21 $\pm$ 0.3	6.50 $\pm$ 2.4	5.86a	75	3.99 $\pm$ 0.5	5.39 $\pm$ 0.6	4.69a
	100	5.27 $\pm$ 0.8	4.76 $\pm$ 1.0	5.02a	100	3.73 $\pm$ 0.5	4.96 $\pm$ 0.5	4.35a
	Mean (SP)	5.65 A	7.12B		Mean (SP)	4.59 A	5.89B	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
PPO ($\text{mmol kg}^{-1} \text{ s}^{-1}$)		1.2	0.6	1.7		1.2	0.9	NS
	0	3.20 $\pm$ 0.2	5.06 $\pm$ 1.2	4.13a	0	5.43 $\pm$ 1.1	4.16 $\pm$ 0.9	4.8a
	50	2.66 $\pm$ 0.6	4.19 $\pm$ 0.6	3.42a	50	5.21 $\pm$ 0.8	5.50 $\pm$ 1.0	5.4a
	75	2.98 $\pm$ 0.8	5.01 $\pm$ 0.4	3.99a	75	5.20 $\pm$ 0.9	5.52 $\pm$ 0.8	5.4a
	100	2.50 $\pm$ 0.5	4.88 $\pm$ 1.0	3.69a	100	4.91 $\pm$ 1.2	5.64 $\pm$ 0.7	5.3a
	Mean (SP)	2.83 A	4.78B		Mean (SP)	5.19 A	5.21 A	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
		NS	0.9	NS		NS	NS	NS

$N = 4$ replicates with 16 fruit per replication. MT = melatonin concentration, SP = storage period, MT $\times$ SP = interaction. LSD = Least Significant Difference at ($P \leq 0.05$). Upper case letters show the statistical difference ($P \leq 0.05$) among the storage durations and lowercase letters indicate the statistical significance among MT treatments and control. Same letter denotes non-significance. The results were expressed as means $\pm$ standard deviation (SD).

the fruit stored for 21 d compared to 28 d (Table 5). The cold storage duration had a significant influence on DHAR activity in both peel and pulp having significantly higher activity in the fruit stored for 21 d compared to 28 d (Table 5). MT ($75 \mu\text{mol L}^{-1}$) significantly increased the activity of enzyme MDHAR in the peel cold stored for 21 d (42.8%) or 28 d (39.9%) compared to the untreated fruit (Table 5). The cold storage duration significantly influenced the activity of MHDAR reporting 47.9% and 68.8% higher activity in the fruit stored for 21 d compared to 28 d in peel and pulp respectively (Table 5).

Experiment 2: Effects of Different Submerge Durations (120, 5 and 3 min) in $75 \mu\text{mol L}^{-1}$ MT on CI in ‘Kensington Pride’ Mango Fruit Cold Stored (5 ± 1 °C) for 21 d

Effects of Different Submersion Periods (120, 5 or 3 min) of MT ($75 \mu\text{mol L}^{-1}$) on CI index and CI Incidence

By the end of the total shelf period of 6 d, all MT (120, 5 and 3 min) submersions showed significantly lower CI index values compared to control showing 40.9%, 38.6%

Table 4 Effects of different concentrations of MT on antioxidant capacity of ‘Kensington Pride’ mangoes cold-stored (5 ± 1 °C) for 21 or 28 d followed by an ambient shelf period of 6 d

Parameter	Peel				Pulp			
	Cold storage and shelf period (d)				Cold storage and shelf period (d)			
SOD($\text{mmol kg}^{-1} \text{ s}^{-1}$)	MT (mmol L^{-1})	21+6	28+6	(Mean MT)	MT (mmol L^{-1})	21+6	28+6	(Mean MT)
	0	0.46 ± 0.04	0.30 ± 0.08	0.38a	0	0.16 ± 0.05	0.04 ± 0.01	0.10a
	50	0.38 ± 0.08	0.43 ± 0.01	0.41a	50	0.19 ± 0.04	0.07 ± 0.01	0.13ab
	75	0.44 ± 0.1	0.37 ± 0.02	0.40a	75	0.23 ± 0.02	0.10 ± 0.02	0.16b
	100	0.54 ± 0.1	0.37 ± 0.04	0.46a	100	0.17 ± 0.05	0.14 ± 0.06	0.15b
	Mean (SP)	0.46B	0.37 A		Mean (SP)	0.19B	0.09 A	
	LSD ($P \leq 0.05$)	MT	SP	MT×SP	LSD ($P \leq 0.05$)	MT	SP	MT×SP
POD ($\text{mmol kg}^{-1} \text{ s}^{-1}$)		NS	0.07	NS		0.022	0.016	NS
	0	2.35 ± 0.42	3.43 ± 0.13	2.89b	0	0.55 ± 0.02	0.59 ± 0.04	0.57a
	50	1.73 ± 0.47	2.64 ± 0.19	2.19a	50	0.52 ± 0.04	0.71 ± 0.08	0.62a
	75	1.87 ± 0.66	3.10 ± 0.21	2.48ab	75	0.57 ± 0.03	0.86 ± 0.24	0.72b
	100	1.91 ± 0.47	2.70 ± 0.19	2.29a	100	0.51 ± 0.06	0.85 ± 0.25	0.68ab
	Mean (SP)	1.97 A	2.96B		Mean (SP)	0.54 A	0.75B	
	LSD ($P \leq 0.05$)	MT	SP	MT×SP	LSD ($P \leq 0.05$)	MT	SP	MT×SP
PAL ($\text{mmol kg}^{-1} \text{ s}^{-1}$)		0.47	0.34	NS		0.07	0.05	NS
	0	0.29 ± 0.05	0.22 ± 0.02	0.25	0	0.09 ± 0.02	0.01 ± 0.007	0.05a
	50	0.25 ± 0.06	0.29 ± 0.01	0.27	50	0.10 ± 0.01	0.15 ± 0.06	0.13b
	75	0.26 ± 0.09	0.26 ± 0.03	0.26	75	0.15 ± 0.01	0.10 ± 0.01	0.12b
	100	0.27 ± 0.06	0.21 ± 0.03	0.24	100	0.04 ± 0.06	0.10 ± 0.02	0.06a
	Mean (SP)	0.27	0.25		Mean (SP)	0.10	0.08	
	LSD ($P \leq 0.05$)	MT	SP	MT×SP	LSD ($P \leq 0.05$)	MT	SP	MT×SP
TPC (g kg^{-1})		NS	NS	NS		0.016	NS	0.02
	0	16.82 ± 0.7	10.87 ± 1.0	13.8a	0	4.330 ± 0.7	3.41 ± 0.8	3.87a
	50	16.09 ± 1.7	10.22 ± 1.1	13.2a	50	6.17 ± 0.9	3.80 ± 1.5	4.99a
	75	20.02 ± 2.0	17.09 ± 1.4	18.6c	75	7.12 ± 0.5	6.09 ± 1.0	6.60b
	100	18.26 ± 1.4	12.68 ± 0.7	15.7b	100	5.38 ± 1.1	4.50 ± 1.1	4.94a
	Mean (SP)	17.79B	12.71 A		Mean (SP)	5.75B	4.45 A	
	LSD ($P \leq 0.05$)	MT	SP	MT×SP	LSD ($P \leq 0.05$)	MT	SP	MT×SP
		0.75	0.53	1.01		1.17	0.83	NS

$N = 4$ replicates with 16 fruit per replication. MT = melatonin concentration, SP = storage period, MT×SP = interaction. LSD = Least Significant Difference at ($P \leq 0.05$). Upper case letters show the statistical difference ($P \leq 0.05$) among the storage durations and lowercase letters indicate the statistical significance among MT treatments and control. Same letter denotes non-significance. The results were expressed as means $\pm$ standard deviation (SD)

and 29.5% reduction in CI index respectively (Fig. 1a). On 21+6 d, MT submersions were able to reduce CI incidence (%) significantly recording 43.4%, 52.2% and 39.1% lesser values in 120-, 5- and 3-min dip treatments respectively compared to control. The lowest CI incidence (%) was recorded in mangoes subjected to 5 min dip treatment (34.4%) while the highest CI incidence (%) was shown in the control (71.9%) fruit (Fig. 1b).

Effects of Different Submersion Periods (120, 5 or 3 min) of MT ($75 \mu\text{mol L}^{-1}$) on SSC, TA, SSC: TA Ratio, Colour and Weight Loss

SSC (%), TA (%) and SSC: TA were significantly influenced by the length of submersion (Table 6). The SSC (%) was significantly lower in the fruit submerged in MT for 120 min compared to 5 min (15.5%) and 3 min (16.3%) submersions

and untreated control (10.9%) (Table 6). TA (%) was significantly lower in the fruit submerged for 5 min compared to 120 min (27.8%) and untreated control (13.3%) (Table 6). The SSC: TA ratio was the highest in the fruit submerged for 5 min in MT compared to 120 min (38.9%) and 3 min (9.5%) treatments and untreated control (17.8%), while it was the lowest in the fruit submerged in MT for 120 min (Table 6). At the end of the cold storage and shelf period (21+6 d), the weight loss (%) was significantly lower in the mangoes submerged in MT compared to untreated control having 14.3%, 17.5% and 14.4% reduction in 120-, 5- and 3-min MT submersions respectively (Fig. 1c). None of the colour parameters were significantly influenced by MT submersions (Table 6).

Table 5 Effects of different concentrations of MT on AsA-GSH cycle in ‘Kensington Pride’ mangoes cold-stored (5 ± 1 °C) for 21 or 28 d followed by an ambient shelf period of 6 d

Parameter	Peel				Pulp			
	Cold storage and shelf period (d)				Cold storage and shelf period (d)			
AsA (mg kg ⁻¹)	MT ($\mu\text{mol L}^{-1}$)	21+6	28+6	(Mean MT)	MT ($\mu\text{mol L}^{-1}$)	21+6	28+6	(Mean MT)
	0	393.0 $\pm$ 54.7	132.0 $\pm$ 28.5	263.0a	0	238.0 $\pm$ 89.4	103.0 $\pm$ 39.1	171.0a
	50	372.0 $\pm$ 155.2	278.0 $\pm$ 60.2	325.0ab	50	379.0 $\pm$ 41.0	138.0 $\pm$ 67.3	258.0a
	75	744.0 $\pm$ 102.5	410.0 $\pm$ 118.8	577c	75	298.0 $\pm$ 47.7	198.0 $\pm$ 106.2	248.0a
	100	483.0 $\pm$ 60.6	343.0 $\pm$ 104.6	413b	100	335.0 $\pm$ 46.0	142.0 $\pm$ 35.3	238.0a
	Mean (SP)	498.0B	291.0 A		Mean (SP)	313.0B	145.0 A	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
GSH (mg kg ⁻¹)		77.8	110.0	NS		NS	56.7	NS
	0	122.3 $\pm$ 31.9	15.1 $\pm$ 5.4	68.7a	0	67.3 $\pm$ 8.0	43.4 $\pm$ 10.3	55.4a
	50	159.8 $\pm$ 18.2	38.0 $\pm$ 10.2	98.9b	50	82.3 $\pm$ 9.8	76.6 $\pm$ 10.5	79.5b
	75	144.8 $\pm$ 24.5	73.3 $\pm$ 2.4	109.1b	75	109.1 $\pm$ 4.4	90.7 $\pm$ 8.7	99.9c
	100	130.5 $\pm$ 34.9	45.2 $\pm$ 19.0	87.9ab	100	91.7 $\pm$ 33.4	56.3 $\pm$ 16.7	74.0ab
	Mean (SP)	139.4B	42.9 A		Mean (SP)	87.6B	66.8 A	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
APX (mmol kg ⁻¹ s ⁻¹)		18.5	26.2	NS		13.6	19.2	27.1
	0	6.82 $\pm$ 1.1	2.39 $\pm$ 0.9	4.61a	0	2.12 $\pm$ 0.4	0.54 $\pm$ 0.1	1.33b
	50	6.65 $\pm$ 1.3	3.88 $\pm$ 0.7	5.26a	50	2.76 $\pm$ 0.2	0.94 $\pm$ 0.2	1.85c
	75	5.97 $\pm$ 2.1	4.95 $\pm$ 0.8	5.46a	75	3.01 $\pm$ 0.2	1.93 $\pm$ 0.2	2.47d
	100	7.42 $\pm$ 2.7	4.71 $\pm$ 1.1	6.06a	100	0.77 $\pm$ 0.4	1.03 $\pm$ 0.3	0.9a
	Mean (SP)	6.71B	3.98 A		Mean (SP)	2.17B	1.11 A	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
GR (mmol kg ⁻¹ s ⁻¹)		NS	1.29	NS		0.34	0.24	0.49
	0	7.75 $\pm$ 0.8	3.29 $\pm$ 0.7	5.52a	0	7.46 $\pm$ 1.1	4.26 $\pm$ 0.8	5.86a
	50	7.28 $\pm$ 1.2	7.93 $\pm$ 1.2	7.60ab	50	11.84 $\pm$ 2.2	6.75 $\pm$ 0.8	9.29b
	75	8.71 $\pm$ 3.5	6.51 $\pm$ 1.0	7.61ab	75	10.38 $\pm$ 0.8	9.27 $\pm$ 0.9	9.82b
	100	9.87 $\pm$ 4.3	7.01 $\pm$ 1.1	8.44b	100	11.14 $\pm$ 3.8	7.30 $\pm$ 2.7	9.22b
	Mean (SP)	8.40B	6.19 A		Mean (SP)	10.20B	6.89 A	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
DHAR (mmol kg ⁻¹ s ⁻¹)		2.39	1.69	NS		2.37	1.68	NS
	0	12.3 $\pm$ 1.0	6.5 $\pm$ 1.0	9.4a	0	5.7 $\pm$ 2.0	3.0 $\pm$ 1.0	4.3a
	50	13.2 $\pm$ 2.0	13.00 $\pm$ 4.0	13.1a	50	7.0 $\pm$ 3.0	3.4 $\pm$ 1.0	5.2a
	75	13.5 $\pm$ 4.0	9.4 $\pm$ 4.0	11.4a	75	9.3 $\pm$ 5.0	7.2 $\pm$ 3.0	8.3a
	100	21.0 $\pm$ 8.0	8.4 $\pm$ 4.0	14.7a	100	8.6 $\pm$ 1.0	8.3 $\pm$ 3.0	6.7a
	Mean (SP)	15.0B	9.4 A		Mean (SP)	7.7B	0.46 A	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
MDHAR (mmol kg ⁻¹ s ⁻¹)		NS	3.7	NS		NS	2.5	NS
	0	3.22 $\pm$ 0.6	2.0 $\pm$ 0.7	2.61a	0	4.10 $\pm$ 1.9	0.98 $\pm$ 0.2	2.54
	50	3.72 $\pm$ 0.8	2.03 $\pm$ 0.7	2.88a	50	5.67 $\pm$ 1.1	1.90 $\pm$ 1.0	3.78
	75	5.63 $\pm$ 2.1	3.33 $\pm$ 0.1	4.48b	75	5.69 $\pm$ 0.5	2.40 $\pm$ 0.3	4.05
	100	3.89 $\pm$ 1.0	1.19 $\pm$ 0.1	2.54a	100	6.06 $\pm$ 1.2	1.46 $\pm$ 0.2	3.76
	Mean (SP)	4.11B	2.14 A		Mean (SP)	5.38B	1.68 A	
	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP	LSD ($P \leq 0.05$)	MT	SP	MT $\times$ SP
		1.23	0.87	NS		NS	0.4	NS

$N = 4$ replicates with 16 fruit per replication. MT = melatonin concentration, SP = storage period, MT $\times$ SP = interaction. LSD = Least Significant Difference at ($P \leq 0.05$). Upper case letters show the statistical difference ($P \leq 0.05$) among the storage durations and lowercase letters indicate the statistical significance among MT treatments and control. Same letter denotes non-significance. The results were expressed as means $\pm$ standard deviation (SD)

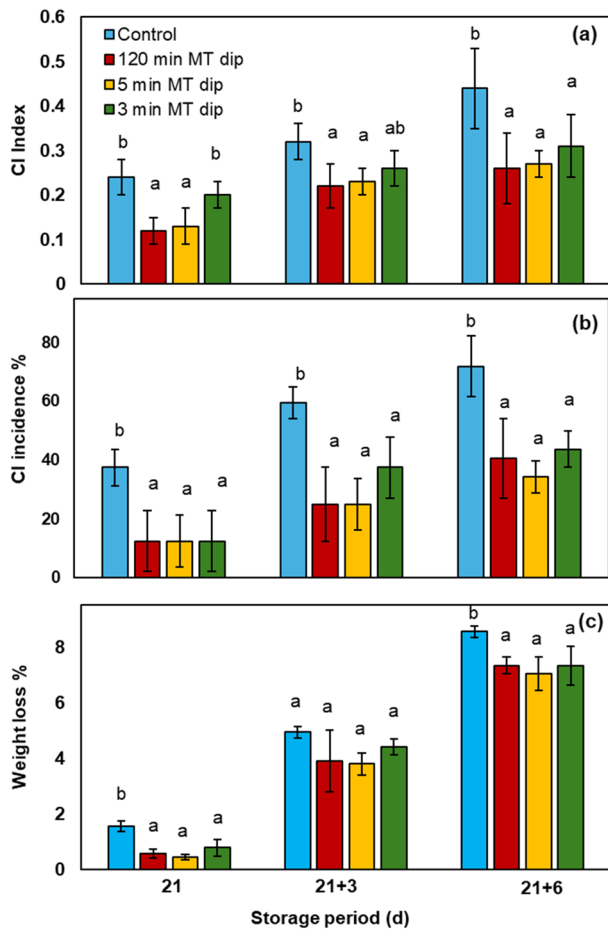


Fig. 1 Effects of different submersion periods in MT ($75 \mu\text{mol L}^{-1}$) on CI index (a), CI incidence % (b) and weight loss % (c). $N = 4$ replicates with 16 fruit per replication. Treatment means significantly different at $P \leq 0.05$ are indicated by different lower-case letters. Same letter denotes non-significance. Standard deviation is represented by vertical bars

Effects of Different Submersion Periods (120, 5 or 3 min) of MT ($75 \mu\text{mol L}^{-1}$) on H_2O_2 , MDA, LOX, PPO and REL in the Peel and the Pulp

MT significantly reduced the levels of H_2O_2 , MDA, and LOX activity in both peel and pulp compared to the untreated fruit (Fig. 2a, b and c). Out of MT treatments, submersion in 120 min showed the highest efficacy in reducing H_2O_2

in the peel and pulp, recording 42.7% and 44.8% reduction respectively (Fig. 2a). Both 120 (45.9%, 33.8%) and 5 min (49.7%, 48.8%) submersions in MT were able to significantly reduce the MDA content in both peel and the pulp compared to control (Fig. 2b). The activity of enzyme LOX in the peel and pulp was significantly lower in fruit submerged in MT for 120 (36.4%, 36.5%), 5 (24.8%, 26.2%) and 3 min (45.3%, 49.0%) than the control (Fig. 2c). MT was not able to affect the activity of enzyme PPO or REL % significantly (Fig. 2d and e).

Effects of Different Submersion Periods (120, 5 or 3 min) of MT ($75 \mu\text{mol L}^{-1}$) on the Performance of Antioxidant Enzymes and TPC

Both 5 min (78.0%) and 3 min (76.0%) submersions in MT significantly increased CAT activity in the peel while, only 3 min (75.5%) submersion was able to increase its activity in the pulp (Fig. 2f). The activity of enzyme SOD was significantly higher in the pulp of the mangoes submerged for 3 min compared to 120 and 5 min (80.0% each) and the untreated control (94.0%) (Fig. 2h). TPC in the peel was significantly higher in the fruit submerged for 120 and 5 min (28.0% and 24.0% respectively) (Fig. 2i). Only 5 min submersion in MT (28.1%) was able to significantly influence the TPC in the pulp compared to control (Fig. 2i).

Effects of Different Submersion Periods (120, 5 or 3 min) of MT ($75 \mu\text{mol L}^{-1}$) on AsA-GSH Cycle

The level of AsA in the peel was significantly higher in the mangoes submerged in MT for 120 and 5 min (67.8%, 61.5% respectively) (Fig. 3a). It was significantly higher in the pulp of mangoes submerged for 120 min (55.3%) compared to control (Fig. 3a). The level of GSH in both peel and pulp was significantly higher in the mangoes submerged in MT for 120 min (52.1%, 65.7% respectively) and 5 min (43.3%, 44.4% respectively) compared to the control (Fig. 3b). The peel APX activity was significantly higher in the fruit submerged in MT for 120 and 5 min (51.7%, 53.3% correspondingly) while all three (120, 5 and 3 min) submersions were able to increase its activity in the

Table 6 Effects of different submersion periods (120, 5 and 3 min) in MT ($75 \mu\text{mol L}^{-1}$) on the quality parameters of 'Kensington Pride' mangoes cold-stored ($5 \pm 1^\circ\text{C}$) for 21 d followed by an ambient shelf period of 6 d

Treatment	Colour parameter					SSC (%)	TA (%)	SSC: TA
	L*	a*	b*	Chroma	Hue angle (h°)			
Control	45.8 $\pm$ 27a	3.0 $\pm$ 1.0a	36.3 $\pm$ 1.6a	36.4 $\pm$ 1.5a	85.3 $\pm$ 1.7a	18.4 $\pm$ 0.8b	0.30 $\pm$ 0.1ab	61.3 $\pm$ 17.6b
75 $\mu\text{mol L}^{-1}$ MT for 120 min	48.4 $\pm$ 1.6a	2.5 $\pm$ 1.2a	37.4 $\pm$ 1.4a	37.5 $\pm$ 1.4a	86.2 $\pm$ 1.9a	16.4 $\pm$ 0.8a	0.36 $\pm$ 0.03b	45.6 $\pm$ 5.3a
75 $\mu\text{mol L}^{-1}$ MT for 5 min	45.6 $\pm$ 2.1a	3.1 $\pm$ 0.3a	38.0 $\pm$ 4.8a	38.2 $\pm$ 4.8a	85.4 $\pm$ 0.8a	19.4 $\pm$ 1.1b	0.26 $\pm$ 0.02a	74.6 $\pm$ 5.8c
75 $\mu\text{mol L}^{-1}$ MT for 3 min	45.6 $\pm$ 2.6a	3.2 $\pm$ 0.4a	38.4 $\pm$ 3.7a	38.6 $\pm$ 3.7a	85.2 $\pm$ 0.5a	19.6 $\pm$ 1.3b	0.29 $\pm$ 0.03ab	67.5 $\pm$ 5.8b

$N = 4$ replicates with 16 fruit per replication. Treatment means significantly different at $P \leq 0.05$ are indicated by different lower-case letters. Same letter denotes non-significance. The results were expressed as means $\pm$ standard deviation (SD)

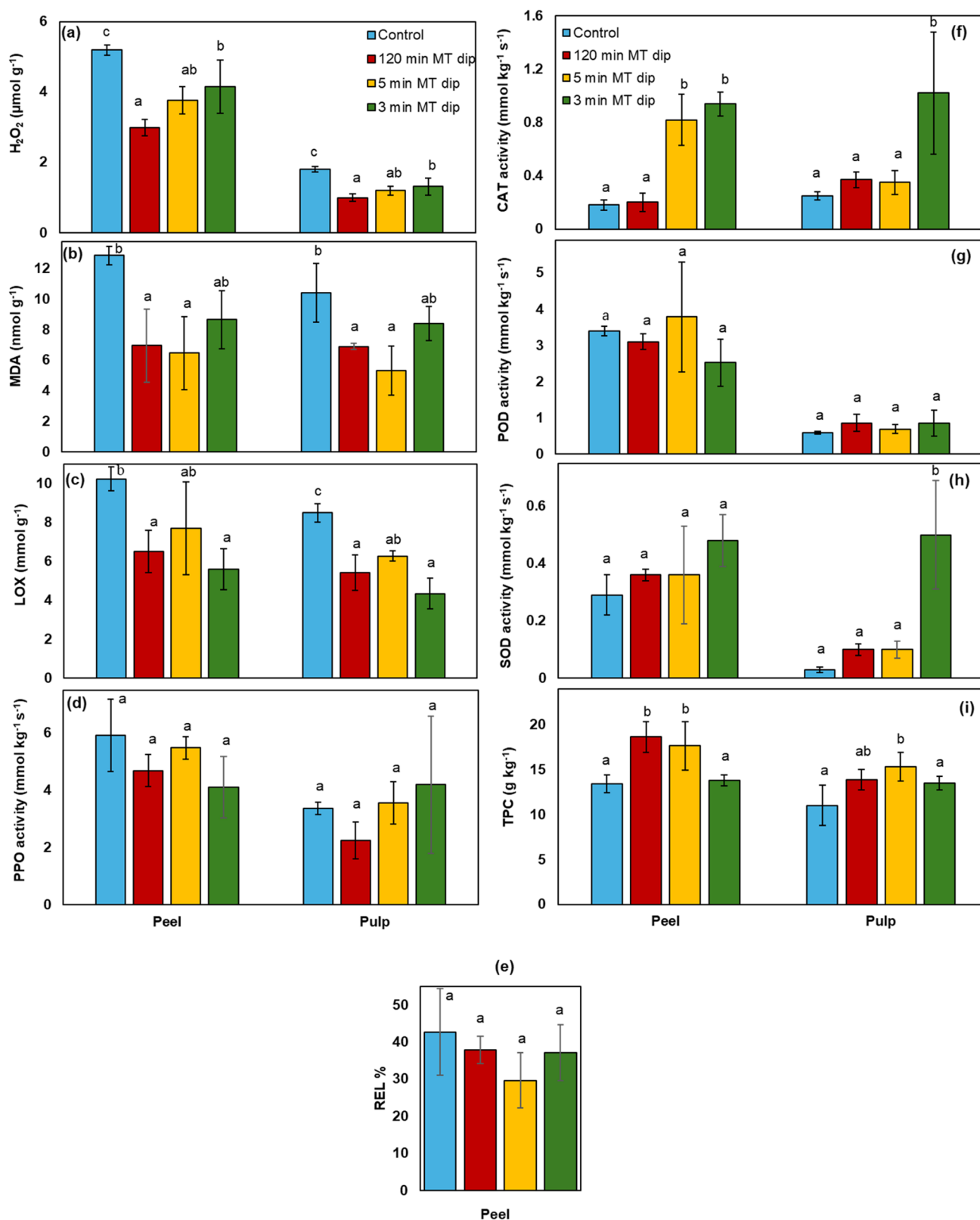


Fig. 2 Effects of different submersion periods in MT ($75 \mu\text{mol L}^{-1}$) on the levels of H₂O₂ (a), MDA (b), LOX (c), PPO (d), REL (e), CAT (f), POD (g), SOD (h), and TPC (i). $N = 4$ replicates with 16 fruit

per replication. Treatment means significantly different at $P \leq 0.05$ are indicated by different lower-case letters. Same letter denotes non-significance. Standard deviation is represented by vertical bars

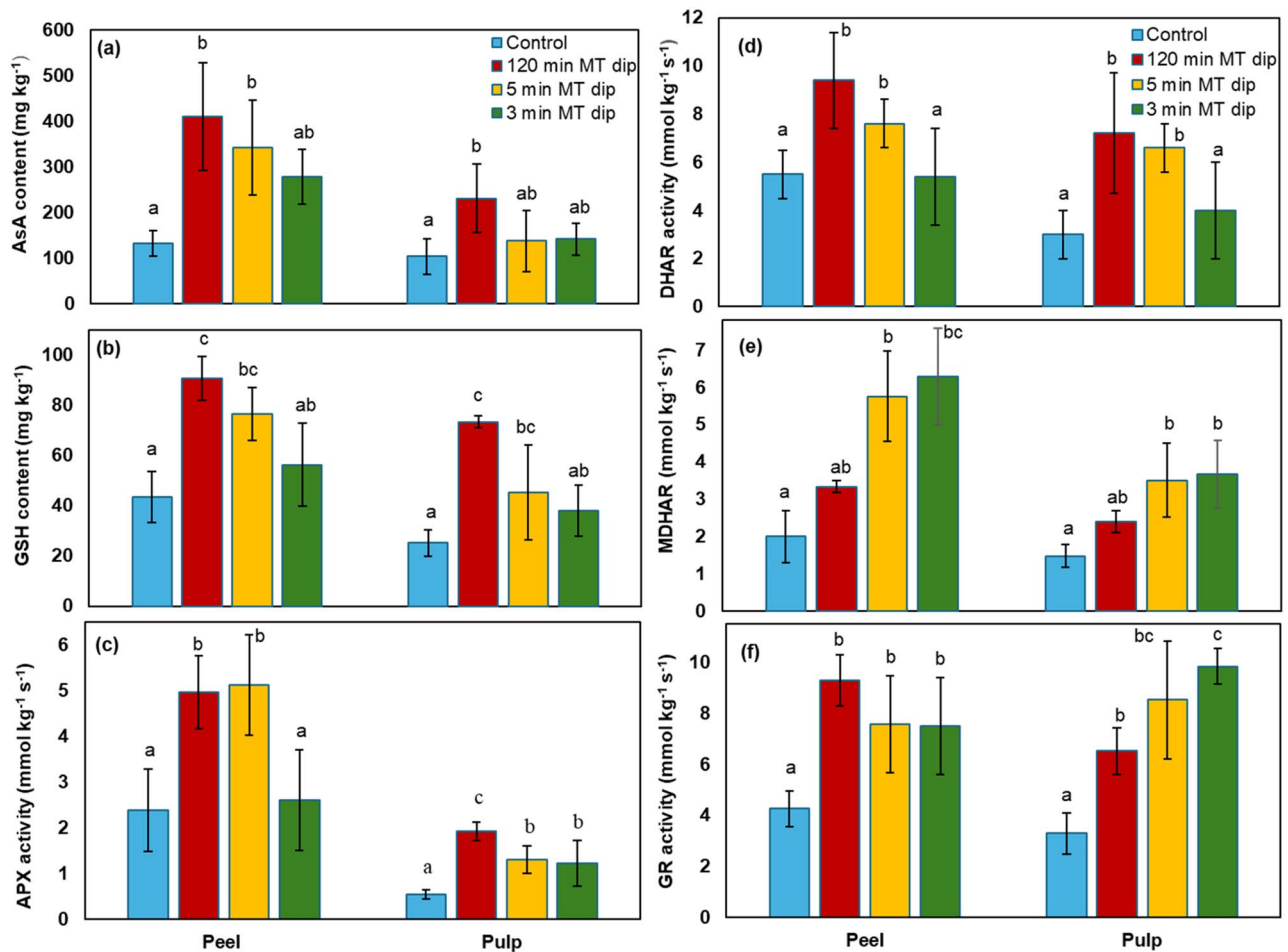


Fig. 3 Effects of different submersion periods of MT (75 µmol L⁻¹) on AsA-GSH cycle. *N* = 4 replicates with 16 fruit per replication. Treatment means significantly different at *P* ≤ 0.05 are indicated by different lower-case letters. Same letter denotes non-significance. Standard deviation is represented by vertical bars

pulp (72.0%, 58.8%, 56.1% respectively) than the control (Fig. 3c). The activity of DHAR in both peel and pulp was significantly higher in the mangoes submerged in MT for 120 min (41.5%, 58.3% respectively) and 5 min (27.6%, 54.5% respectively) compared to untreated fruit (Fig. 3d). MDHAR activity was significantly increased by both 5 min (65.3%, 57.8%) and 3 min (68.2%, 59.7%) MT submersions in the peel and pulp compared to control correspondingly (Fig. 3e). The activity of GR was significantly increased by all three MT submersions (120, 5 and 3 min) in both peel and pulp compared to control (Fig. 3f). The increase in the peel was 54.0%, 43.7%, and 43.0%, while it was recorded as 49.5%, 61.4%, and 66.5% in the pulp correspondingly compared to untreated mangoes.

ent lower-case letters. Same letter denotes non-significance. Standard deviation is represented by vertical bars

Discussion

CI is the major obstacle when storing mangoes at low temperatures. Noticeably darkened lenticels and slightly wrinkled greyish areas on the skin were gradually increased during the 6-d shelf period when 'Kensington Pride' mangoes were removed from cold storage (5 ± 1 °C for 21 or 28 d). In agreement, Sati et al. (2025) stated that the prominence of lenticels due to cold-induced phenolic compound accumulation around them, is the first visible symptom of CI in mangoes. The sunken greyish areas and pitting on the peel appears with the progression of CI due to loss of membrane integrity and other metabolic and histological changes (Sati et al. 2025). In the presence of oxygen, polyphenols are oxidised by PPO leading to the production of brown - black pigments. In this study, significantly lower CI incidence and CI index (Table 1; Fig. 1a and b) were observed in the fruit immersed in 75 µmol L⁻¹ MT compared to the control,

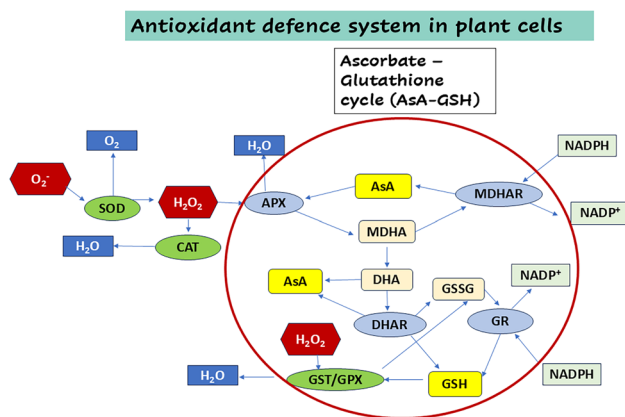


Fig. 4 Antioxidant defence system in plant cells (Hasanuzzaman et al. 2020)

50 and 100 $\mu\text{mol L}^{-1}$ MT. Hormesis, which causes a low dose stimulation, and a high dose inhibition (Calabrese and Agathokleous 2023) could possibly be the reason for better effectivity of 75 $\mu\text{mol L}^{-1}$ compared to 100 $\mu\text{mol L}^{-1}$ in reducing CI. Previously, nitric oxide induced hormesis was able to inhibit the biotic and abiotic stress damage in plants (Calabrese and Agathokleous 2023). According to Duarte-Sierra et al. (2020), abiotic stress hormesis is extensively used to extend postharvest storage life and quality of fruits and vegetables. However, MT induced hormetic effects on CI in mango fruit should be further investigated.

At cellular level, the lack of membrane integrity is the first indication of CI (Aghdam and Bodbodak 2014). The accumulation of reactive oxygen species (ROS) such as H_2O_2 leads to lipid peroxidation. This is accelerated by LOX activity, leading to the production of MDA, further inducing membrane damage (Li et al. 2023; Sanches et al. 2022). Additionally, direct membrane rupture due to cold stress results in leakage of cellular materials and water, causing cell death (Tomita et al. 2023). Thus, cell membrane damage is often assessed using the levels of H_2O_2 , MDA, REL and the activities of LOX and PPO in cold stored fruit (Li et al. 2023; Sanches et al. 2022). To minimise such oxidative stress, different defence mechanisms such as ROS avoidance and detoxification are adopted by cellular mitochondria (Fig. 4). Hence, as a way of ROS detoxification, the production of antioxidant enzymes (CAT, PAL, POD, SOD) is triggered in cold-stressed tissues (Song et al. 2016). In addition to above antioxidant enzymes, AsA-GSH cycle (Fig. 4) provides a vital pathway for cellular ROS detoxification (Hasanuzzaman et al. 2020). Hence, the significantly lower CI incidence and CI index observed in this study could be a collective result of significantly reduced H_2O_2 , MDA, REL, LOX activity and increased activities of SOD, POD, PAL and the TPC content in both peel and pulp (Table 3; Fig. 2). The influence of MT on reducing CI in ‘Kensington

Pride’ mangoes was found to be concentration dependent, 75 $\mu\text{mol L}^{-1}$ showing the best results.

A similar observation was reported in ‘Langra’, ‘Chausa’, ‘Dashehari’ ‘Keitt’ and Guifei mangoes as well (Bhardwaj et al. 2022b). Among Indian cultivars, ‘Langra’ showed the highest responsiveness to MT (100 $\mu\text{mol L}^{-1}$ for 120 min) with minimum CI, recording lowest levels of H_2O_2 , MDA, LOX activity as well as the highest levels of TPC, and highest activities of PAL, CAT, SOD, POD and APX (Bhardwaj et al. 2022b). ‘Guifei’ mangoes treated with MT (500 $\mu\text{mol L}^{-1}$ for 60 min) reported reduced levels of REL, MDA and LOX activity resulting in significantly lower CI compared to untreated fruit (Xu et al. 2023).

Additionally, MT was not capable of reducing CI, when the ‘Kensington Pride mangoes were stored for 28 d at low temperature. This observation could be a result of significantly higher levels of H_2O_2 , MDA, REL, LOX and PPO with significantly lower levels of SOD, POD, TPC, AsA, GSH, APX, GR, DHAR, and MDHAR in the mangoes stored for 28 d compared to those stored for only 21 d. This significant influence of storage duration in MT treated fruits seems to be cultivar dependent, as in contrast to our observations, MT was able to reduce CI in ‘Langra’ and ‘Chausa’ up to 28 d and in Guifei mangoes up to 40 d (Xu et al. 2023).

Significantly higher chilling tolerance observed in MT treated cultivar ‘Langra’ was also ascribed to higher accumulation of endogenous polyamines, γ -aminobutyric acid (Bhardwaj et al. 2022b) and higher proline levels (Bhardwaj et al. 2021) compared to non-responsive cultivar ‘Gulab Jamun’. Probably, the differences in the levels of these compounds in ‘Kensington Pride’ mangoes, which were not analysed in the present study, would have contributed to its lower response to MT compared to ‘Langra’ and ‘Guifei’ mangoes.

However, an accelerated deterioration was observed in Indian cultivars after 21 d of cold storage, while cultivar ‘Gulab Jamun’ was not responsive at all to any MT treatment showing CI symptoms by 7th d of cold storage (Bhardwaj et al. 2022b).

When quality parameters were considered, the weight loss which primarily occurs due to transpiration and respiratory loss of moisture was not significantly influenced under experiment 1 (Table 1), while it was significantly lower compared to untreated mangoes under experiment 2 (Fig. 1). The weight loss in Indian mangoes was also not affected by MT treatments (Bhardwaj et al. 2021). The reason for the reduction in weight loss under experiment 2 in the mangoes harvested two months later (in late autumn) could be attributed to differences in temperature during growth, leading to higher assimilate reserves, but this needs further investigation. In experiment 2, TA % was significantly lower in

the fruit submerged for 5 min making SSC: TA ratio significantly higher (Table 6). A decrease in TA is possibly due to using up of organic acids during cellular respiration (Bhardwaj et al. 2021). However, TA was not influenced by MT under experiment 1 (Table 2), probably due to the differences in growing temperatures and corresponding assimilate reserves.

In the present study, the colour of 'Kensington Pride' mangoes was not influenced by MT, but colour coordinates were differently influenced by the storage duration (Tables 2 and 6). In contrast, the development of colour was significantly delayed in 'Guifei' mangoes immersed in 500 $\mu\text{mol L}^{-1}$ MT for 1 h before cold storage (Liu et al. 2020).

Even though the influence on CI was not assessed in their study, Rastegar et al. (2020) found a significantly positive impact on the quality of mango fruit submerged for 10 min in MT before storage at 15 °C, reporting higher CAT and POD activities as well as reduced PPO. Due to these encouraging results, quick dips (5 and 3 min) in MT (75 $\mu\text{mol L}^{-1}$) were trialled in this study to improve the practicality of MT application. Interestingly, there was not any significant difference observed in CI index, CI incidence and weight loss among 120-, 5- and 3-min MT immersions, all three treatments having significantly lower values compared to untreated mangoes (Fig. 1). This could be attributed to similar influence on the levels and activities of H_2O_2 , MDA, LOX, PPO, TPC, AsA, DHAR, MDHAR and GR showed by both 120- and 5-min immersions and significantly higher activities observed in CAT, SOD, GR, MDHAR and APX in the mangoes submerged for 3 min (Figs. 1, 2 and 3). Thus, shorter contact time might be optimal for MT absorption. Prolonged immersion may have caused additional stress by inducing physiological responses such as anaerobic respiration. Another reason might be the influence of exposure duration on the uptake kinetics of MT. The uptake kinetics of MT through mango peels or other fruit surfaces are yet to report. However, cellular studies have revealed that glucose transporters GLUT/SLC2A and oligopeptide transporter PEPT1/2 are involved in MT uptake, and it is substantially influenced by the level of glucose (Mayo et al. 2017). Possible anaerobic respiration during prolonged submersion might have caused a reduction in the level of glucose adversely influencing MT uptake by mitochondria. Anaerobic respiration could be the reason for significantly lower level of SSC recorded in mangoes dipped for 120 min compared to shorter submersions (Table 6). However, further studies focused on uptake kinetics of MT are required to comprehend the reason behind better or equivalent performance of 5 min MT dip treatment compared to the 120 min MT dip treatment in this study.

Positive response of 'Kensington Pride' mangoes to a significantly shorter contact time could also be attributed

to its significantly lower rate of ethylene production during ripening compared to previously studied cultivars. At the climacteric peak of ethylene production, 'Kensington Pride' mangoes produce around 0.007 $\text{pmol kg}^{-1} \text{s}^{-1}$ under ambient ripening (Vithana et al. 2018b). However, 'Langra' (0.03 $\text{nmol kg}^{-1} \text{s}^{-1}$), 'Chausa' (0.07 $\text{nmol kg}^{-1} \text{s}^{-1}$), 'Dashehari' (0.05 $\text{nmol kg}^{-1} \text{s}^{-1}$) and 'Gulab Jamun' (0.12 $\text{nmol kg}^{-1} \text{s}^{-1}$) mangoes produce significantly higher rates of ethylene at the climacteric peak of ripening even during cold storage (Bhardwaj et al. 2021). Similarly, 'Guifei' mangoes (0.5 $\text{nmol kg}^{-1} \text{s}^{-1}$) also produce a significantly higher rate of ethylene during ripening at cold storage (Liu et al. 2020). Additionally, the dynamics of endogenous polyamines, γ -aminobutyric acid and proline levels of 'Kensington Pride' mangoes subjected to MT treatments and cold storage also need to be analysed in future studies to further explore any cultivar influence.

Conclusion

MT at the concentration of 75 $\mu\text{mol L}^{-1}$ was the most effective in reducing CI in 'Kensington Pride' mango fruit cold stored at 5 ± 1 °C for up to 21 d. The immersion periods 120, 5–3 min in 75 $\mu\text{mol L}^{-1}$ MT exhibited significant reduction in the CI index and CI incidence. Improved membrane stability achieved through reduced LOX and PPO activities, and the improved antioxidant potential acquired by intensified activities of antioxidant enzymes CAT, PAL, SOD, POD as well as the better performance of AsA-GSH cycle in detoxifying ROS in MT-treated mangoes may have contributed to this reduction in CI up to 21 d cold storage. Colour, SSC and TA were not largely impacted by MT submersion. Overall, 5 min submersion of mango fruit in MT showed the highest efficacy. Hence, this study for the first time revealed that 5 min submersion in 75 $\mu\text{mol L}^{-1}$ MT solution is sufficient to reduce CI in 'Kensington Pride' mangoes stored at 5 ± 1 °C up to 21 d followed by 6 d ripening on shelf. Although a significant reduction was observed under 75 $\mu\text{mol L}^{-1}$ MT submersion, MT was unable to eliminate CI in 'Kensington Pride' mangoes cold stored for 21 d, recording 40.6% (120 min) and 34.4% (5 min) CI incidence by the end of the shelf period. Thus, future studies on the synergistic effect of MT in combination with other promising compounds in reducing CI in mangoes warrant investigation. Gene expression, endogenous melatonin quantification and signalling validation in a future study would further explain the mode of action of MT in mitigating CI in 'Kensington Pride' mangoes. Additionally, to explore the full adoption potential of MT 5 min dip treatment, a cost-benefit analysis and a workflow integration study with wastewater

management should be carried out in future with different mango cultivars grown under diverse conditions.

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Declarations

Competing Interests The authors have no competing interests to declare that are relevant to the content of this article.

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