

Research paper

**Enhancement of salt tolerance in quinoa (*Chenopodium quinoa* var. *Titicaca*)
by seed priming with melatonin**

Manizhe Jahantighi¹, Parto Roshandel^{2*}, and Abdolrazagh Danesh Shahraki¹

Received: January 16, 2023 Accepted: June 12, 2023

¹Department of Agronomy, Faculty of Agriculture, Shahrekord University, Shahrekord, Iran

²Department of Biology, Faculty of Basic Sciences, Shahrekord University, Shahrekord, Iran

*Corresponding author; Email: roshandelparto@gmail.com

Abstract

In plants, melatonin (N-acetyl-5-methoxytryptamine) contributes to various environmental stress responses and developmental processes. Accordingly, the effect of seed priming with melatonin (1250 μM) on increasing salt tolerance at 40 dS m^{-1} NaCl was studied in *Chenopodium quinoa* var. *Titicaca* (quinoa) using a factorial experiment based on a completely randomized design at Shahrekord University in 2019. In the present experiment, different levels of NaCl and melatonin were first applied and according to the obtained results, 1250 μM melatonin and 40 dS m^{-1} NaCl were selected for the main study. The measured traits and indices included the fresh and dry weight, the content of photosynthetic pigments, hydrogen peroxide (H_2O_2), malondialdehyde (MDA), as well as the activity of catalase, ascorbate peroxidase, and guaiacol peroxidase. Seed priming with melatonin reduced the adverse effects of NaCl in the salt-stressed quinoa plants by increasing the dry weight (two-fold) and the level of photosynthetic pigments (1.2-fold) compared to the salinity stress alone. Also, the application of melatonin by changing the activity of antioxidant enzymes, reduced the content of H_2O_2 by 25% and MDA by 42%, which resulted in the reduction of oxidative stress in the quinoa plants under salinity conditions. Overall, it can be concluded that seed priming with an optimal level of melatonin can be a proper technique to increase salt tolerance in quinoa under highly saline conditions by reducing the harmful effects of salinity-induced oxidative stress by reducing the H_2O_2 and lipid peroxidation levels. It also protects the photosynthetic machinery by reducing the degradation of photosynthetic pigments which leads to the increased growth of the quinoa plants under salinity conditions.

Keywords: oxidative stress, quinoa, reactive oxygen species (ROS), salinity, seed pretreatment

How to cite: Jahantighi M, Roshandel P, Danesh Shahraki A. 2023. Seed priming with melatonin enhances salt tolerance in quinoa (*Chenopodium quinoa* var. *Titicaca*). J Plant Physiol Breed. 13(2): 181-195.

Introduction

In recent decades the reduction of freshwater resources due to climate change and decrease in rainfall, and also, the increase in the reduction of agricultural fields caused by soil salinization, demands new strategies for the sustainability of food supply. The economic benefits of halophytic species for widespread use in arid and semi-arid areas and the application of techniques to increase salt

tolerance in these species are of interest.

In recent years, it has been reported that more than one billion hectares of the world's land have been affected by soil salinity, and this destructive event continues to expand day by day (Tian *et al.* 2020; Hopmans *et al.* 2021). It is said that the amount of soil salinity worldwide is more than 2 Mha per year (Singh 2018). Currently, soil salinity and the problems caused by it have spread to more than 100

countries (Hammam and Mohamed 2020).

Plants are damaged by salinity through water stress, ion toxicity, and nutritional imbalance. Furthermore, salinity stress in plants leads to oxidative stress, i.e. overproduction of ROS, which causes damage to vital macromolecules such as proteins and nucleic acids, and cellular membranes (Shi *et al.* 2015).

Although seed priming is recognized as a useful technique for improving seed germination and establishment of emergence uniformity, many reports have shown that it can also support safer seedling growth under stressful conditions (Rasheed *et al.* 2022).

Melatonin, as an indoleamine, has a similar function to auxin. Also, melatonin acts as an antioxidant as well as a molecule in signaling transduction pathways and affects various physiological processes of different organisms including bacteria, fungi, invertebrates, vertebrates, algae, and plants (Zhan *et al.* 2019). This chemical acts directly to scavenge ROS produced by environmental stressors. Moreover, melatonin is considered a candidate plant hormone with important roles and also is involved in plant responses to a variety of biotic and abiotic stresses such as salinity.

Several studies including seed priming or foliar application of melatonin, have shown the important and vital role of melatonin in increasing salt tolerance in different plant

species such as *Triticum aestivum* (Ke *et al.* 2018), *Vicia faba* (Dawood *et al.* 2015), *Solanum lycopersicum* (Martinez *et al.* 2018), *Oryza sativa* (Liang *et al.* 2015), *Zea mays* (Chen *et al.* 2018), *Raphanus sativus* (Yao *et al.* 2017), *Cucumis sativus* (Zhang *et al.* 2017), *Helianthus annuus* (Arora and Bhatla, 2017). Moreover, it was revealed that melatonin treatment increased salt and drought tolerance in soybean (*Glycine max*) plants and improved their yield (Wei *et al.* 2015). A similar result was observed in Bermuda grass (*Cynodon dactylon*), so exogenous melatonin treatment induced cold, drought, and salt tolerance in this species compared to untreated plants (Shi *et al.* 2015). Likewise, the mitigation effects of melatonin treatment have been reported in other plants such as *Citrus aurantium* (Kostopoulou *et al.* 2015), *Arabidopsis thaliana* (Chen *et al.* 2017), and *Malus hupehensis* (Gong *et al.* 2017) under salinity conditions.

Quinoa (*Chenopodium quinoa*) is a herbaceous annual plant belonging to Amaranthaceae. Its edible seeds are rich in vitamins (A, B₁, B₂, B₉, and E) and minerals (calcium, magnesium, iron, copper, zinc, and lithium), essential amino acids (even greater than in many grains), and are a valuable source of carbohydrates and essential fatty acids for human nutrition (Repo-Carrasco *et al.* 2003; Hariadi *et al.* 2011).

Eisa *et al.* (2012) noted that *Chenopodium*

quinoa would be considered a promising salt-tolerant species since it can be successfully cultivated in low to medium concentrations of NaCl (up to 40% seawater salinity). As a cash-crop halophyte, quinoa grows even better in moderate salinity (100 mM) than in non-saline conditions (Adolf *et al.* 2013). The resistance of quinoa to environmental stresses along with genetic diversity has made it a suitable candidate for planting in different and harsh agro-climatic regions of Iran.

In the current research, the effect of seed priming with melatonin on increasing salt tolerance in quinoa (*Chenopodium quinoa* var. Titicaca) under high NaCl levels was investigated. Also, the possible protective effects of melatonin by managing oxidative stress under saline conditions were studied.

Materials and Methods

Plant material and preparation

Seeds of quinoa (*Chenopodium quinoa* var. Titicaca) were obtained from, Karaj, Iran. They were surface sterilized with 70% ethanol for 2 min followed by repeated washing with double-distilled water and dried on the filter paper. The experiment was conducted at Shahrekord University in 2019. Our preliminary experiment was conducted at the germination stage with seven levels of salinity including 0, 10, 20, 30, 40, 50, and 60 dS.m⁻¹ NaCl and seven levels (0.50, 100, 500, 750, 1000, and 1250 µM) of melatonin solution

(dissolved in distilled water). The seeds were soaked in melatonin (Sigma, Germany) for 8 h at 25 ± 1 °C under dark conditions. After determining the most efficient concentration of melatonin (1250 µM) at the germination stage, we conducted an experiment to evaluate the effect of melatonin priming with the 1250 µM concentration on the growth of 70-day-old quinoa plants. We also applied 40 dS m⁻¹ NaCl (the concentration that reduced seed germination of quinoa by 50%). Salinity treatment was applied to the 10-day-old seedlings of quinoa for two months in a greenhouse (16 h light/8 h dark, 35/26 °C temperature, with 65% humidity). The treatments comprised (i) control (no treatment), (ii) seed priming with 1250 µM melatonin, (iii) 40 dS m⁻¹ NaCl as saline conditions (no priming), (iv) seed priming with 1250 µM melatonin plus 40 dS m⁻¹ NaCl. Treated and untreated seeds (16 seeds per pot) from which seedlings were reduced to 10 at the onset of salt treatment) were planted in polystyrene pots (Diameter 32 cm / Height 24 cm), containing 50% soil and 50% fine sand (Shams 2012). Then, at the onset of the salt treatment, the number of seedlings was reduced to 10.

To obtain the constant level of sodium chloride in the soil, the pots were irrigated with an appropriate amount of water to wash the sodium chloride from the previous irrigation, and then the new saline solution was added to

the pot.

Samples were taken from the 70-day-old plants to measure dry weight, lipid peroxidation, photosynthetic pigments content, and antioxidant enzymes in quinoa plants. Dry weight was measured after drying plants for five days at 70 °C until they reached a constant weight.

Assessment of photosynthetic pigments

The content of total chlorophyll and carotenoids was measured based on the method of Lichtenthaler and Buschmann (2001) with 80% acetone as the solvent:

$$\text{Chlorophyll a } (\mu\text{g ml}^{-1}) = 12.25A_{663} - 2.79A_{645}$$

$$\text{Chlorophyll b } (\mu\text{g ml}^{-1}) = 21.5A_{645} - 5.1A_{663}$$

$$\text{Total Chlorophyll } (\mu\text{g ml}^{-1}) = \text{Chlorophyll (a + b)}$$

$$\text{Carotenoids} = [1000A_{470} - (1.82)(\text{Chlorophyll a}) - (85.02)(\text{Chlorophyll b})]/198$$

where, A_{663} , A_{645} , and A_{470} represent absorbance values read at 663, 645, and 470 nm wavelengths, respectively.

Assessment of lipid peroxidation

Lipid peroxidation was assessed by measuring the malondialdehyde (MDA) content in the shoots of quinoa plants based on the method of Ksouri *et al.* (2007). The extinction coefficient was $155 \text{ mM}^{-1} \text{ cm}^{-1}$.

Measuring H₂O₂ content

The concentration of H₂O₂ in the shoots was

measured using the titanium-hydroperoxide complex (Nag *et al.* 2000). The absorbance of the orange-yellow H₂O₂-Ti complex was determined at 410 nm against blank. The H₂O₂ content was obtained through a standard curve plotted with the known concentration of H₂O₂ in the range of 10 to 100 μM .

Enzyme extraction and assay

The enzyme extraction was based on the method of Chen (2000) with some modifications. All the following operations were carried out at 4 °C. The extract was kept in the -20 °C freezer. The catalase (CAT) activity was measured by a spectrophotometer by determining the H₂O₂ consumption (Extinction Coefficient = $39.4 \text{ mM}^{-1} \text{ cm}^{-1}$) at 240 nm in 50 mM phosphate buffer, pH 7.5, and 200 mM H₂O₂ (Nemat-Ala and Hassan 2006). The activity of ascorbate peroxidase (APX) was measured by a spectrophotometer according to the method of Kato and Shimizu (1985) at 280 nm in 0.2 mM potassium phosphate buffer, pH 7.5, 15 mM ascorbic acid, and 50 mM H₂O₂, as ascorbate (Extinction coefficient = $2.8 \text{ mM}^{-1} \text{ cm}^{-1}$) was oxidized. The activity of glutathione peroxidase (GPX) was assessed in 44 mM H₂O₂ and 45 mM guaiacol. The absorption at 470 nm was read and the activity was calculated using the extinction coefficient of $26.6 \text{ mM}^{-1} \text{ cm}^{-1}$ (Buchanan and Balm 2005).

Statistical analysis

The experiments were conducted as a completely randomized design with three replications. The data were analyzed by SAS (V. 9.0) software. *P*-values less than 0.05 were defined as statistically significant.

Results

The results of the analysis of variance for the

physiological and biochemical characteristics of quinoa plants under saline conditions and priming with melatonin are shown in Table 1. Salinity and priming with melatonin had a significant effect on all of the studied traits ($p \leq 0.01$). Besides, a significant interaction between melatonin priming and salinity was obtained ($p \leq 0.01$).

Salinity (40 dS m⁻¹ NaCl) decreased the

Table 1. Analysis of variance for some physiological and biochemical traits of the 70-day-old plants of quinoa primed with melatonin and grown under saline conditions.

SOV	Mean squares				
	df	Dry weight	Fresh weight	Total chlorophyll	Carotenoids
Melatonin priming	1	0.574**	1.952**	5.619**	12.724**
Salinity	1	1.333**	4.89**	9.580**	32.107**
Melatonin priming × Salinity	1	0.484**	1.81**	3.329**	0.856**
Error	8	0.002	0.008	0.051	0.008
CV (%)	-	2.73	2.72	2.43	1.42

** : Significant at $p \leq 0.01$.

Table 1 continued

SOV	Mean squares					
	df	H ₂ O ₂	MDA	CAT	APX	GPX
Melatonin-priming	1	6.331**	0.009**	0.323**	0.122**	0.013**
Salinity	1	12.448**	0.403**	0.257**	4.869**	0.028**
Melatonin-priming × Salinity	1	26.542**	0.001**	0.039**	0.460**	0.166**
Error	8	13.447	0.00003	0.001	0.001	0.0003
CV (%)	-	18.32	4.241	1.54	5.463	0.0003

** : Significant at $p \leq 0.01$; MDA: Malondialdehyde; CAT: Catalase; APX: Ascorbate peroxidase; GPX: Glutathione peroxidase

fresh weight of quinoa plants by 2.6 times (from 3.95 to 1.52 g) compared to the control ($p \leq 0.01$) (Figure 1A). However, melatonin priming at 1250 μ M compensated for this reduction under salt stress by two-fold (from 1.52 to 3.08 g) ($p \leq 0.01$). Under normal conditions, priming with melatonin had no significant effect on the fresh weight.

The dry weight of the quinoa plants

decreased by 60.6% (from 2.08 to 0.82 g) under 40 dS m⁻¹ NaCl ($p \leq 0.01$) (Figure 1B). However, priming with melatonin (1250 μ M) significantly compensated this reduction of salt-stressed plants by almost two-fold (from 0.82 to 1.7 g). Under normal conditions, the priming did not have a significant effect on this characteristic.

Salt stress at the concentration of 40 dS

m^{-1} NaCl decreased the total chlorophyll content in quinoa by 26.5% (from 3.83 to 2.81 $\mu\text{g ml}^{-1}$) ($p \leq 0.01$) (Figure 2A). However, at 40 dS m^{-1} NaCl, priming with melatonin increased total chlorophyll content by 1.2-fold (from 2.81 to 3.32 $\mu\text{g ml}^{-1}$). In the non-stressed quinoa plants, melatonin priming (1250 μM) increased the total chlorophyll content by 1.7-fold compared to the control (from 3.83 to 6.51 $\mu\text{g ml}^{-1}$).

In the absence of salinity, melatonin priming (1250 μM) caused an increase (1.8-

fold; from 3.34 to 6 $\mu\text{g ml}^{-1}$) in carotenoids content of the quinoa plants ($p \leq 0.01$) (Figure 2B). Salinity (40 dS m^{-1} NaCl) decreased the level of carotenoids by 45% (from 3.34 to 1.84 $\mu\text{g ml}^{-1}$). However, the use of priming with melatonin (1250 μM) increased the carotenoid content significantly (1.2-fold; from 1.84 to 2.2 $\mu\text{g ml}^{-1}$) ($p \leq 0.01$) (Figure 2B).

The results showed that the H_2O_2 content in the salinized shoots of quinoa increased by 1.5-fold (from 3.8 to 5.87 $\mu\text{Mg}^{-1}\text{fr.wt}$) compared to the control ($p \leq 0.01$) (Figure 3A).

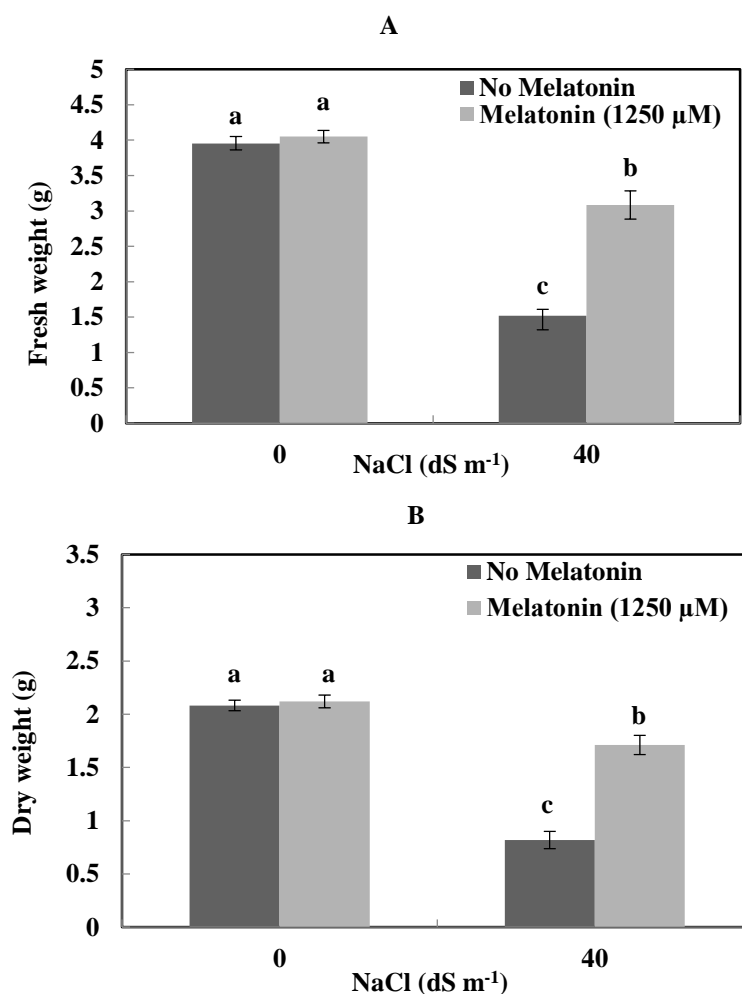


Figure 1. Fresh weight (A) and Dry weight (B) of the 70-day-old quinoa plants from unprimed or primed seeds with melatonin (1250 μM) and grown under normal or saline conditions (40 dS m^{-1} NaCl). The means with the different letter(s) are significantly different at $p \leq 0.01$ based on the LSD test. Bars indicate \pm standard error.

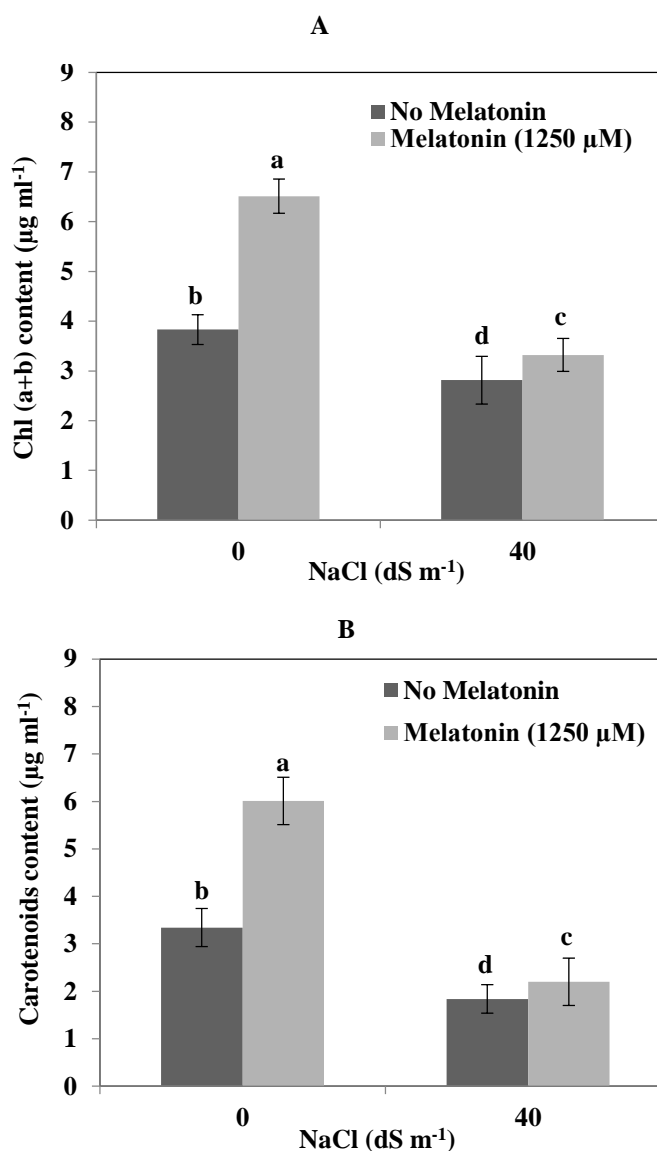


Figure 2. Total chlorophyll content (A) and (B) carotenoid content of 70-day-old quinoa plants from unprimed or primed seeds with melatonin (1250 μM) and grown under normal or saline conditions (40 dS m⁻¹). The means with the different letter(s) are significantly different at $p \leq 0.01$ based on the LSD test. Bars indicate \pm standard error.

Priming with melatonin (1250 μM) reduced the H_2O_2 level by 25% in the salinized shoots (from 5.87 to 4.4 $\mu\text{Mg}^{-1}\text{fr.wt}$). In the absence of salinity, the application of melatonin priming decreased the H_2O_2 content by 13.5% (from 3.8 to 3.3 $\mu\text{Mg}^{-1}\text{fr.wt}$) in the quinoa plants ($p \leq 0.01$) (Figure 3A).

Salinity (40 dS m⁻¹ NaCl) increased the MDA level (as a marker of lipid peroxidation) in the shoots of quinoa plants by 2.5-fold (from 0.075 to 0.191 mmol MDA g⁻¹ fr.wt.) compared to the control ($p \leq 0.01$) (Figure 3B). Nevertheless, melatonin priming (1250 μM) significantly reduced the level of lipid

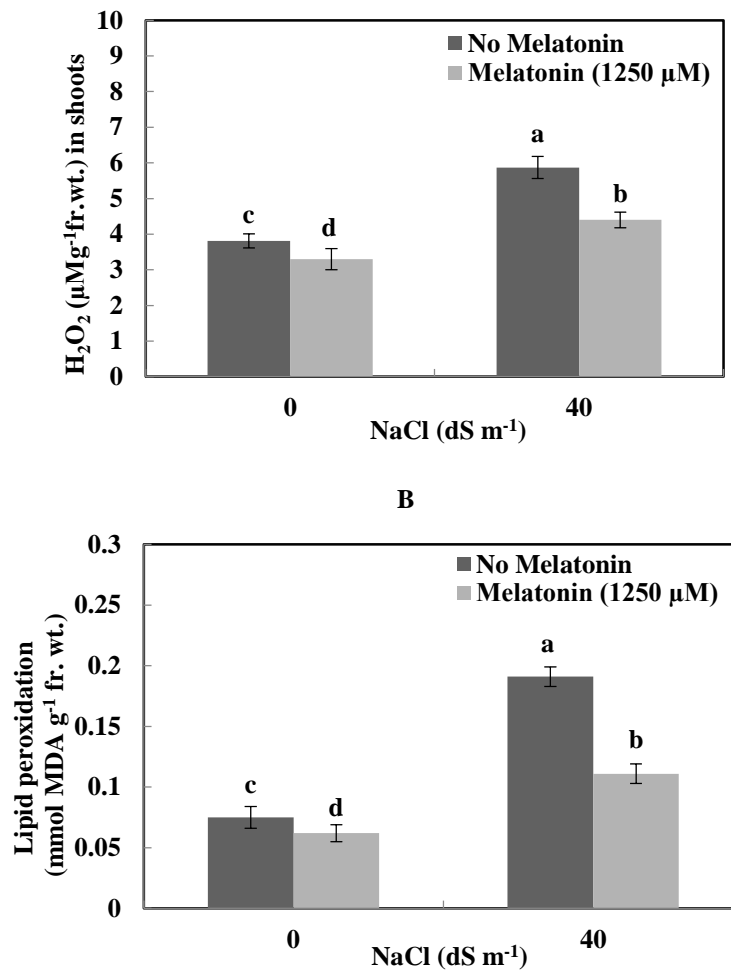


Figure 3. The H₂O₂ content (A) and malondialdehyde (MDA) content of the 70-day-old quinoa plants from unprimed or primed seeds with melatonin (1250 μM) and grown under normal or saline conditions (40 dS m⁻¹). The means with the different letter(s) are significantly different at $p \leq 0.01$ based on the LSD test. Bars indicate \pm standard error.

peroxidation of the cellular membrane by 42% in the salt-stressed quinoa plants (from 0.191 to 0.111 fr. wt.) ($p \leq 0.01$). Furthermore, in the absence of salinity, melatonin priming caused a 17% decrease (from 0.075 to 0.062) in the MDA level in the quinoa plants compared to the control.

Salt stress (40 dS m⁻¹ NaCl) caused a significant increase (1.2-fold) in the CAT activity in the quinoa shoots ($p \leq 0.01$) (Figure 4A). However, priming with melatonin (1250 μM) caused a 16% decrease in the CAT

activity under salinity conditions. Under normal conditions (0 mM NaCl), CAT activity decreased by 1.23-fold in the primed plants ($p \leq 0.01$).

Salt stress (40 dS m⁻¹ NaCl) caused an increase (1.3-fold) in the APX activity compared to the control ($p \leq 0.01$) (Figure 4B). However, the melatonin priming significantly stressed quinoa plants compared to the salinity treatment alone. In the absence of NaCl, melatonin priming significantly increased the activity of APX compared to the control ($p \leq$

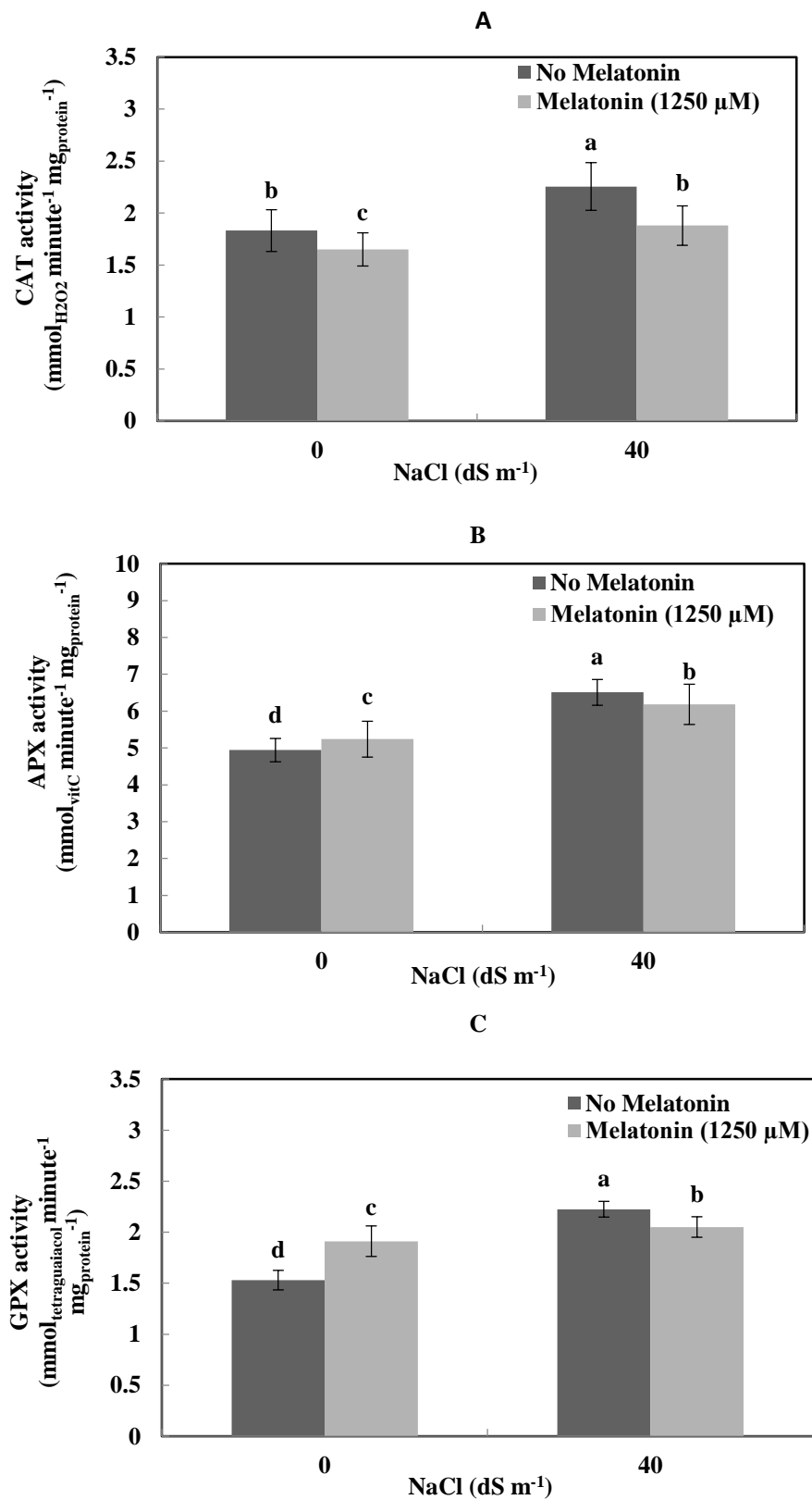


Figure 4. The catalase (CAT) activity (A), ascorbate peroxidase (APX) activity (B), and glutathione peroxidase (GPX) activity (C) of the 70-day-old quinoa plants from unprimed or primed seeds with melatonin (1250 μM) and grown under normal or saline conditions (40 dS m⁻¹). The means with the different letter(s) are significantly different at $p \leq 0.01$ based on the LSD test. Bars indicate \pm standard error.

0.01) (Figure 4B).

As shown in Figure 4C, the activity of GPX exhibited an increase (up to 1.5-fold) in shoots of quinoa plants under saline conditions as compared to the control. However, priming with melatonin under salinity conditions caused a decrease (7%) in the activity of this enzyme compared to the salinity alone. Under normal conditions, priming with melatonin caused an augmentation in the GPX activity by 1.24-fold compared to the control.

Discussion

Our results showed that seed priming with the optimum level of melatonin (1250 μM) improved the quantity of biomass, membrane integrity, and content of photosynthetic pigments, and decreased the H_2O_2 level in the quinoa plants compared with the control under salinity conditions. These findings are in agreement with the previous works which have indicated that melatonin-treated plants would be more salt tolerant with lower levels of ROS and lipid peroxidation (Arnao and Hernández-Ruiz 2015; Chen *et al.* 2018). Also, melatonin-treated plants have exhibited a greater height and biomass compared to untreated plants (Li *et al.* 2019).

Salt stress not only constrains different aspects of plant physiology but also induces the overproduction of ROS (such as H_2O_2) leading to oxidative stress (Li *et al.* 2019).

High levels of ROS result in DNA damage, protein denaturation, lipid peroxidation in cellular membranes, pigment breakdown, carbohydrate oxidation, and impaired enzyme activities. Thus, plants must sustain a balance between ROS production and ROS scavenging under stress. Regarding to ROS scavenging, plants have two systems of antioxidants to lessen damage due to oxidative stress: the enzymatic and non-enzymatic systems. Antioxidant enzyme systems include superoxide dismutase, guaiacol peroxidase, CAT, and APX. The non-enzymatic system (including carotenoids, ascorbic acid, tocopherols, glutathione, and phenolic compounds) is also crucial for ROS removal. As suggested, phyto-melatonin is involved in clearing excessive ROS and consequently reducing the injurious effects of saline conditions in plants. Accordingly, melatonin enhances plant tolerance to salinity by reducing oxidative stress in two ways. First, melatonin acts as an antioxidant and directly contributes to clear ROS. Melatonin's capacity to scavenge ROS is said to be greater than the common antioxidants such as vitamin C and vitamin E (Sun *et al.* 2021). Second, melatonin manages indirect pathways such as inducing the activity of antioxidant enzymes, the performance of photosynthesis, metabolite levels, ion homeostasis, as well as the metabolism of phytohormones. Furthermore,

melatonin can induce the expression of genes involved in the tolerance of plants to stresses (Li *et al.* 2019).

Under salt stress, the antioxidant content of enzymes generally increases and their activities depend on the level of plant salt tolerance (Guo *et al.* 2018). In our experiment also, the activity of measured antioxidant enzymes exhibited an increment in the quinoa plants under salinity stress. Besides, in line with the previous studies (Zhang *et al.* 2017; Li *et al.* 2019), melatonin pretreatment modified the activity of antioxidant enzymes (e.g. increasing the activity of APX and GPX) in quinoa plants under normal conditions.

It has been suggested that melatonin can prevent oxidative damage by reducing free radicals and reducing electron leakage from the electron transport chain in the mitochondria. Under environmental stresses, melatonin frequently up-regulates the content of antioxidant enzymes in plants by regulating antioxidant-related gene expression (Nawaz *et al.* 2018). It was suggested that melatonin protects plants against abiotic stress by activating the ascorbate-glutathion cycle which is an important antioxidant system in higher plants. As found in the soybean plant, exogenously applied melatonin is permeable across the membranes and increases the endogenous melatonin content, leading to improved plant growth and grain yield (Wei *et al.* 2015).

In addition to the antioxidant effects of melatonin, this chemical also participates in protecting plant photosynthesis under salt stress. Photosynthesis is very sensitive to environmental stresses such as drought and salinity and is usually damaged when faced with these stresses. Melatonin can protect photosynthesis by increasing chlorophyll content, electron transport chains, and stomatal conductance to reduce photosynthetic inhibition caused by stress (Hernández-Ruiz and Arnao, 2018). Melatonin protects plant chlorophyll against various stresses by down-regulating the chlorophyll degradation genes (Zuo *et al.* 2017). The use of melatonin also regulates the electron transport system, such as improving nonphotochemical quenching or photochemical quenching, and the maximal quantum yield of PSII photochemistry (Fleta-Soriano *et al.* 2017). Consistent with the findings of other studies, the current work showed that melatonin reduces chlorophyll degradation in quinoa plants under saline conditions.

The optimum level of exogenously applied melatonin is species-specific. For example, in *Arabidopsis* seedlings, a low concentration of melatonin (10–20 μM) has no obvious effect, a moderate concentration (40 μM) promotes plant growth and development, and a high concentration (200–400 μM) prevents plant growth (Bajwa *et al.* 2014). In

the present research, the most efficient level of melatonin for seed priming was 1250 μM .

Conclusion

Overall, it can be concluded that seed priming with melatonin (1250 μM) can be a useful method to promote salt tolerance in *Chenopodium quinoa* var. Titicaca (quinoa) under highly saline conditions. Melatonin increased salt tolerance in the quinoa by reducing the injurious effects of oxidative stress due to salinity by reducing H_2O_2 and

lipid peroxidation levels in the quinoa. Moreover, melatonin can protect the photosynthetic machinery by reducing the chlorophyll degradation caused by salt stress which leads to increased growth of quinoa under high NaCl levels.

Conflict of interest

The authors declare that they have no conflict of interest with any organization concerning the subject of the manuscript.

References

- Adolf VI, Jacobsen SE, Shabala S. 2013. Salt tolerance mechanisms in quinoa (*Chenopodium quinoa* Willd.). *Environ Exp Bot.* 92: 43-54.
- Arnao MB, Hernández-Ruiz J. 2015. Functions of melatonin in plants: a review. *J Pineal Res.* 59(2): 133-150.
- Arora D, Bhatla SC. 2017. Melatonin and nitric oxide regulate sunflower seedling growth under salt stress accompanying differential expression of Cu/Zn SOD and Mn SOD. *Free Radic Biol Med.* 106: 315-328.
- Bajwa VS, Shukla MR, Sherif SM, Murch SJ, Saxena PK. 2014. Role of melatonin in alleviating cold stress in *Arabidopsis thaliana*. *J Pineal Res.* 56(3): 238-245.
- Buchanan BB, Balmer Y. 2005. Redox regulation: a broadening horizon. *Annu Rev Plant Biol.* 56: 187-220.
- Chen DH, Ye HC, Li GF. 2000. Expression of a chimeric farnesyl diphosphate synthase gene in *Artemisia annua* L. transgenic plants via *Agrobacterium tumefaciens*-mediated transformation. *Plant Sci.* 155(2): 179-185.
- Chen Z, Xie Y, Gu Q, Zhao G, Zhang Y, Cui W, Xu S, Wang R, Shen W. 2017. The AtrbohF-dependent regulation of ROS signaling is required for melatonin-induced salinity tolerance in *Arabidopsis*. *Free Radic Biol Med.* 108: 465-477.
- Chen YE, Mao JJ, Sun LQ, Huang B, Ding CB, Gu Y, Liao JQ, Hu C, Zhang ZW, Yuan S, *et al.* 2018. Exogenous melatonin enhances salt stress tolerance in maize seedlings by improving antioxidant and photosynthetic capacity. *Physiol Plant.* 164(3): 349-363.
- Dawood MG, El-Awadi ME. 2015. Alleviation of salinity stress on *Vicia faba* L. plants via seed priming with melatonin. *Acta Biolo Colomb.* 20(2): 223-235.
- Eisa S, Hussin S, Geissler N, Koyro HW. 2012. Effect of NaCl salinity on water relations, photosynthesis and chemical composition of Quinoa (*'Chenopodium quinoa'* Willd.) as a potential cash crop halophyte. *Aust J Crop Sci.* 6(2): 357-368.
- Fleta-Soriano E, Díaz L, Bonet E, Munné-Bosch S. 2017. Melatonin may exert a protective role against drought stress in maize. *J Agron Crop Sci.* 203(4): 286-294.

- Gong X, Shi S, Dou F, Song Y, Ma F. 2017. Exogenous melatonin alleviates alkaline stress in *Malus hupehensis* Rehd. by regulating the biosynthesis of polyamines. *Molecules*. 22(9): 1542.
- Guo YY, Tian SS, Liu SS, Wang WQ, Sui N. 2018. Energy dissipation and antioxidant enzyme system protect photosystem II of sweet sorghum under drought stress. *Photosynthetica*. 56(3): 861-872.
- Hammam AA, Mohamed ES. 2020. Mapping soil salinity in the East Nile Delta using several methodological approaches of salinity assessment. *Egypt J Remote Sens Space Sci*. 23: 125-131.
- Hariadi Y, Marandon K, Tian Y, Jacobsen SE, Shabala S. 2011. Ionic and osmotic relations in quinoa (*Chenopodium quinoa* Willd.) plants grown at various salinity levels. *J Exp Bot*. 62(1): 185-193.
- Hernández-Ruiz J, Arnao MB. 2018. Relationship of melatonin and salicylic acid in biotic/abiotic plant stress responses. *Agronomy*. 8(4): 33
- Hopmans JW, Qureshi AS, Kisekka I, Munns R, Grattan SR, Rengasamy P, Ben-Gal A, Assouline S, Javaux M, Minhas PS, *et al.* 2021. Critical knowledge gaps and research priorities in global soil salinity. *Adv Agron*. 169: 1-191.
- Kato M, Shimizu S. 1985. Chlorophyll metabolism in higher plants. VI. Involvement of peroxidase in chlorophyll degradation. *Plant Cell Physiol*. 26(7): 1291-1301.
- Ke Q, Ye J, Wang B, Ren J, Yin L, Deng X, Wang S. 2018. Melatonin mitigates salt stress in wheat seedlings by modulating polyamine metabolism. *Front Plant Sci*. 9: 914.
- Kostopoulou Z, Therios I, Roumeliotis E, Kanellis AK, Molassiotis A. 2015. Melatonin combined with ascorbic acid provides salt adaptation in *Citrus aurantium* L. seedlings. *Plant Physiol Biochem*. 86: 155-165.
- Ksouri R, Megdiche W, Debez A, Falleh H, Grignon C, Abdely C. 2007. Salinity effects on polyphenol content and antioxidant activities in leaves of the halophyte *Cakile maritima*. *Plant Physiol Biochem*. 45(3-4): 244-249.
- Li J, Liu J, Zhu T, Zhao C, Li L, and Chen M, 2019. The role of melatonin in salt stress responses. *Int J Mol Sci*. 20(7): 1735.
- Liang C, Zheng G, Li W, Wang Y, Hu B, Wang H, Wu H, Qian Y, Zhu XG, Tan DX, *et al.* 2015. Melatonin delays leaf senescence and enhances salt stress tolerance in rice. *J Pineal Res*. 59(1): 91-101.
- Lichtenthaler HK, Buschmann C. 2001. Chlorophylls and carotenoids: measurement and characterization by UV-VIS spectroscopy. *Curr Protoc Food Anal Chem*. 1(1): F4.3.1- F4.3.8.
- Martinez V, Nieves-Cordones M, Lopez-Delacalle M, Rodenas R, Mestre TC, Garcia-Sanchez F, Rubio F, Nortes PA, Mittler R, Rivero RM. 2018. Tolerance to stress combination in tomato plants: new insights in the protective role of melatonin. *Molecules*. 23(3): 535.
- Nag S, Saha K, Choudhuri MA. 2000. A rapid and sensitive assay method for measuring amine oxidase based on hydrogen peroxide–titanium complex formation. *Plant Sci*. 157(2): 157-163.
- Nawaz MA, Jiao Y, Chen C, Shireen F, Zheng Z, Imtiaz M, Bie Z, Huang Y. 2018. Melatonin pretreatment improves vanadium stress tolerance of watermelon seedlings by reducing vanadium concentration in the leaves and regulating melatonin biosynthesis and antioxidant-related gene expression. *J Plant Physiol*. 1(220): 115-127.
- Nemat-Ala MM, Hassan NM. 2006. Changes of antioxidants levels in two maize lines following atrazine treatments. *Plant Physiol Biochem*. 44(4): 202-210.
- Rasheed R, Ashraf MA, Hussain I, Ali S, Riaz M, Iqbal M, Farooq U, Qureshi FF. 2022. Priming effect in developing abiotic stress tolerance in cereals through metabolome reprogramming. In: Roychoudhury A, Aftab T, Acharya K. (eds.) *Omics approach to manage abiotic stress in cereals*. Singapore: Springer. pp. 47-71.
- Repo-Carrasco R, Espinoza C, Jacobsen SE. 2003. Nutritional value and use of the Andean crops quinoa (*Chenopodium quinoa*) and kañiwa (*Chenopodium pallidicaule*). *Food Rev Int*. 19(1-2): 179-189.

- Shams AS. 2012. Response of quinoa to nitrogen fertilizer rates under sandy soil conditions. In Proceedings of 13th International Conference of Agronomy, Faculty of Agriculture, Benha University, 9-10 September, Egypt.
- Shi H, Jiang C, Ye T, Tan DX, Reiter RJ, Zhang H, Liu R, Chan Z. 2015. Comparative physiological, metabolomic, and transcriptomic analyses reveal mechanisms of improved abiotic stress resistance in bermudagrass [*Cynodon dactylon* (L). Pers.] by exogenous melatonin. *J Exp Bot.* 66(3): 681-694.
- Singh A. 2018. Alternative management options for irrigation induced salinization and waterlogging under different climatic conditions. *Ecol Indic.* 90: 184-192.
- Sun C, Liu L, Wang L, Li B, Jin C, Lin X. 2021. Melatonin: a master regulator of plant development and stress responses. *J Integr Plant Biol.* 63(1): 126- 145.
- Tian F, Hou M, Qiu Y, Zhang T, Yuan Y. 2020. Salinity stress effects on transpiration and plant growth under different salinity soil levels based on thermal infrared remote (TIR) technique. *Geoderma.* 357: 113961.
- Wei W, Li QT, Chu YN, Reiter RJ, Yu XM, Zhu DH, Zhang WK, Ma B, Lin Q, Zhang JS, *et al.* 2015. Melatonin enhances plant growth and abiotic stress tolerance in soybean plants. *J Exp Bot.* 66(3): 695-707.
- Yao H, Wang X, Lin L. 2017. Effects of melatonin treated radish on the growth of following stubble lettuce under salt stress. In: 4th International Conference on Renewable Energy and Environmental Technology. Atlantis Press. pp. 387-390.
- Zhan H, Nie X, Zhang T, Li S, Wang X, Du X, Tong W, Song W. 2019. Melatonin: a small molecule but important for salt stress tolerance in plants. *Int J Mol Sci.* 20(3): 709.
- Zhang J, Zeng B, Mao Y, Kong X, Wang X, Yang Y, Zhang J, Xu J, Rengel Z, Chen Q. 2017. Melatonin alleviates aluminum toxicity through modulating antioxidative enzymes and enhancing organic acid anion exudation in soybean. *Funct Plant Biol.* 44(10): 961-968.
- Zhang N, Zhang HJ, Sun QQ, Cao YY, Li X, Zhao B, Wu P, Guo YD. 2017. Proteomic analysis reveals a role of melatonin in promoting cucumber seed germination under high salinity by regulating energy production. *Sci Rep.* 7(1): 1-15.
- Zuo Z, Sun L, Wang T, Miao P, Zhu X, Liu S, Song F, Mao H, Li X. 2017. Melatonin improves the photosynthetic carbon assimilation and antioxidant capacity in wheat exposed to nano-ZnO stress. *Molecules.* 22(10): 1727.

افزایش تحمل به شوری در کینووا (*Chenopodium quinoa* var. Titicaca)**توسط پرایمینگ بذر با ملاتونین****منیژه جهان تیغی^۱، پرتو روشندل^{۲*} و عبدالرزاق دانش شهرکی^۱**

۱- گروه زراعت، دانشکده کشاورزی، دانشگاه شهرکرد، شهرکرد

۲- گروه زیست شناسی، دانشکده علوم پایه، دانشگاه شهرکرد، شهرکرد

مسئول مکاتبه: Email: roshandelparto@gmail.com

چکیده

در گیاهان، ملاتونین (N-استیل-۵-متوکسی تریپتامین) در پاسخ به تنش‌های مختلف محیطی و فرایندهای نموی مشارکت می‌نماید. بر این اساس، تاثیر پرایمینگ بذر با ملاتونین (۱۲۵۰ میکرومولار) روی افزایش تحمل به شوری (۴۰ دسی زیمنس بر متر کلرید سدیم) در کینووا (*Chenopodium quinoa* var. Titicaca) با استفاده از آزمایش فاکتوریل بر پایه طرح کاملاً تصادفی با سه تکرار در دانشگاه شهرکرد در سال ۱۳۹۸ مورد بررسی قرار گرفت. در این تحقیق، ابتدا سطوح مختلف کلرید سدیم و ملاتونین به کار گرفته شد و بر اساس نتایج به دست آمده، ملاتونین با غلظت ۱۲۵۰ میکرومولار و کلرید سدیم با غلظت ۴۰ دسی زیمنس بر متر برای آزمایش اصلی انتخاب گردید. صفات و شاخص‌های اندازه گیری شده عبارت بودند از وزن تر و خشک، میزان رنگیزه‌های فتوسنتزی، پراکسید هیدروژن (H_2O_2)، مالون دآلدئید (MDA)، و نیز فعالیت کاتالاز، آسکوربات پراکسیداز و گایاکول پراکسیداز. پرایمینگ بذر با ملاتونین با افزایش وزن خشک (۲ برابر) و سطح رنگدانه‌های فتوسنتزی (۱/۲ برابر) در مقایسه با تنش شوری به تنهایی، اثرات نامطلوب NaCl را در گیاهان کینووا تحت تنش شوری کاهش داد. همچنین کاربرد ملاتونین با تغییر فعالیت آنزیم‌های آنتی اکسیدانی، کاهش میزان H_2O_2 (۲۵ درصد) و MDA (۴۲ درصد) باعث کاهش تنش اکسیداتیو در گیاهان تحت تنش کینووا شد. به طور کلی، می‌توان نتیجه گرفت که پرایمینگ بذر با سطح بهینه ملاتونین می‌تواند تکنیک مناسبی برای افزایش تحمل به شوری در کینووا تحت شرایط بسیار شور باشد و این کار از طریق کاهش اثرات مضر تنش اکسیداتیو ناشی از شوری یعنی کاهش سطح H_2O_2 و MDA انجام می‌گیرد. همچنین ملاتونین با کاهش تخریب رنگدانه‌های فتوسنتزی، از دستگاه فتوسنتزی محافظت می‌کند و این امر منجر به افزایش رشد گیاهان کینووا تحت تنش شوری می‌شود.

واژه‌های کلیدی: انواع اکسیژن فعال، پیش تیمار بذر، تنش اکسیداتیو، شوری، کینووا