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H_2O_2 SEED PRIMING TO ALLEVIATE THE SALINITY EFFECT IN TOMATO (*SOLANUM LYCOPERSICON*)

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Abstract

Salinity is one of the important constraints of sustainable agriculture, globally reducing crop production by impairing various biochemical, physiological and molecular functions. To examine hydrogen peroxide (H_2O_2) role to palliate the salinity stress, we investigated the seed-inducing effect of different H_2O_2 concentrations (0, 50, 100 and 200 mM) in tomato (*Solanumlycopersicon*) crops resistance to salt stress. During this study, we estimated the eventuality of preparing tomato seeds with H_2O_2 to promote seed sprouting, plant growth and induce salt stress tolerance. Soaked seeds were pre-germinated and grown in pots on different saline soils (0, 40 and 80 mMNaCl). Findings revealed that saltness explosively affected tomato germination and seedling growth. Seed germination with 50 and 100 mM H_2O_2 improved germination in saline solutions, while high levels of hydrogen peroxide pretreatment (200 mM) inhibited tomato germination. In the pot experiment, application of hydrogen peroxide significantly increased tomato fresh weight (85% at 200 mM dose) compared to controls. At 80 mMNaCl, the concentrations 50 and 100 mM of H_2O_2 areshowing the most important effect on tomato fresh weight. Results indicate that treatments have a positive effect on membrane integrity (100% at 200 mM) and a general positive effect was observed for all properties measured under salt stress. At the same time, H_2O_2 priming significantly increased catalase activity. Seed preparation with hydrogen peroxide could be an efficient sustainable way to reduce tomato salinity stress.

Keys words: Seed priming, Hydrogen peroxide, Salt stress, Tomato, Germination, Plant growth, Plant physiology, Catalase.

Introduction

The impact of salt stress on food security has become a significant threat in the agricultural sector. The increasing salinity levels due to climate change are significantly impacting food production on new farmlands (Hussain *et al*., 2018). In fact, soil salinity disrupts various plant physiological processes, leading to oxidative damage and alterations in leaf gas exchange, ultimately resulting in decreased plant growth (Trușcă *et al*., 2023). High salinity levels also disrupt plant metabolism by increasing ion toxicity, reducing the availability of essential elements and affecting the synthesis of proteins and lipids (Houmani *et al*., 2022). This leads to premature plant senescence, reduced photosynthetic efficiency, and ultimately decreased crop yields. Plants have developed mechanisms to detect external signals and respond effectively to stress. When plants encounter abiotic and biotic stressors, there is an increase in the production of reactive oxygen species (ROS), leading to oxidative damage to cellular structures and plant molecules. Among them, we can mention the hydrogen peroxide, which a stable molecule that can freely diffuse. It plays a crucial role in voltage signaling pathways (Andrés *et al*., 2022). It can therefore stimulate a number of adaptive responses that enhance resistance to biotic and abiotic stressors. Antioxidant systems that adjust the hydrogen peroxide levels encompass of two types of H_2O_2 scavengers: enzymatic including ascorbate peroxidase, peroxidoxin, catalase and glutathione reductase as well as non-enzymatic compounds like flavonoids, glutathione and ascorbate continuously regulate ROS concentrations in H_2O_2(Martemucci *et al.*, 2022). Futhermore, plants have continuously developing a manner to use small amount titers of ROS as a signaling component to maintain different processes to response to environmentally stress (Tarkowski *et al.*, 2022). At higher concentrations, it contributes cell metabolites oxidative damage. But otherwise, at low concentrations it generates cell signaling (Fujii *et al.*, 2022). In particular, Redox imbalances affiliated with abiotic stress like salinity increase metabolic

rates and ultimately regulate H_2O_2 production in plant cells (Trușcă *et al.*, 2023). Over the last decade, the latest studies have revealed that pretreatment with suitable H_2O_2 amount by expressing specifics and controlling different stress-responsive pathways can enhance the abiotic stress tolerance (Sachdev *et al.*, 2021).

The small size and comparatively lengthy lifetime of H_2O_2 molecules lets them to pass via cell membranes to different cell compartment making easy various signaling function (Hasanuzzaman *et al.*, 2020). Therefore, priming is considered the greatest mechanism to induce stress tolerance in plants (Fujii *et al.*, 2022), and increasing evidence indicates that initial exposure to chemical conditioning agents makes plants more resistant to stress (Hönig *et al.*, 2023). Priming has been proved as good as a modulator for abiotic stress tolerance (Salam *et al.*, 2022). Subsequently, plants utilize an antioxidant enzymatic system to repair damage caused by FAOx. For example, superoxide dismutase (SOD) helps neutralize the O_2^- anion by converting it into H_2O_2, which can be harmful to the plant. Other enzymes, such as catalases (CAT) and ascorbate peroxidase (APX), are then utilized to transform H_2O_2 into water and oxygen. This process acts as a stress signal for the plant, prompting it to activate its antioxidant system (Hasanuzzaman *et al*., 2020). Although plants able to make a spacious range of antioxidants that can mitigate oxidative damage, other exogenous applications must be further investigated, including preparing seeds with stressors or chemical mixture that make them extra proof against subsequent stress situations.

Priming is considered the greatest mechanism to induce stress tolerance in plants (Fujii *et al.*, 2022), and increasing evidence indicates that initial exposure to chemical conditioning agents makes plants more resistant to stress (Hönig *et al.*, 2023). In additionaly, priming has been proved as good as a modulator for abiotic stress tolerance (Salam *et al.*, 2022). This process acts as a stress signal for the plant, prompting it to activate its antioxidant system (Hasanuzzaman *et al*., 2020). Despite priming importance from a point of view agronomic and ecologic, the fundamental mechanisms worried in processing this plant are still unknown (Tripathi *et al*., 2024). In fact, understanding the mechanisms behind stress tolerance induced by hydrogen peroxide is valuable for identifying biotechnological approaches to improve resistance to abiotic stress. Maintaining a precise balance between H_2O_2 secretion and depletion is crucial to using H_2O_2 as a signaling molecule without causing toxicity. To evaluate the effects of H_2O_2 priming, experiments were conducted on tomato plants exposed to salinity stress. The tomato *Solanum Lycopersicon* is a highly important market garden crop globally and plays a strategic role in Tunisian agriculture, covering 13.7% of total area dedicated to market gardening. It is a popular vegetable widely consumed by both urban and rural Tunisians. However, Tunisia's heavy reliance on surface water for irrigation puts it at risk of water shortages due to changing weather patterns caused by the climate crisis (Mkaddem *et al*., 2022). Additionally, the tomato's supply is constrained by low yields caused by various biotic and abiotic factors, including salinity. Tomatoes are known to be sensitive to salt levels and are one of the most important vegetables worldwide (Agius *et al*., 2022). In this study, we examined the effects of different levels of hydrogen peroxide on reducing the negative impact of soil salinity during germination. We also looked at how H_2O_2 priming can induce changes in morphology, biochemistry, and physiology to reduce the consequences of oxidative and osmotic stress caused by salinity in tomato plants.

Material and Methods

Plant material: In all the experiments, tomato seeds of the "Rio Grande" variety were used.

Seed priming with H_2O_2: Seed priming is a method that involves hydrated seeds tokickstart the germination process and activating pre-germination metabolic functions. This technique was suggested as a way to improve seed vigor before planting. During this study, tomato seeds were subjected to varying concentrations of hydrogen peroxide solutions (0, 50, 100, and 200 mM) in a dark environment at 22°C for 2 hours. They were then rinsed with distilled water to remove any excess chemicals and further cleansed with a 40% sodium hypochlorite solution for 15 minutes, followed by 3 washes with sterile distilled water.

Germination experiment: Seed germination was carried out in Murashige and Skoog Medium (Murashige & SKoog, 1962) using plastic Petri dishes. After priming period, the seeds were salinized by adding salt stress-inducing agent NaCl at various concentrations: 0, 40, 80 and 120 mM. We performed an experiment 4x4 factorial organized in randomized layout (4 salt concentrations: 0, 40, 80, and 120 Mm NaCl) and 4 concentrations of hydrogen peroxide priming (0, 50, 100 and 100 mM). Ten tomato seeds were placed in each of the 48 Petri dishes, 10 seedlings were arbitrarily selected from each Petri dish during germination period and stem length turned into measured in cm.

Germination was evaluated every 48 h and percentage germination (GP) was determined:

$$GP = \frac{\text{Number of normally germinated seeds}}{\text{Total number of seeds sown}} \times 100$$

Plant growth: An experiment was performed under greenhouse conditions. Temperature varied between 20°C on night time and 25°C on day, relative humidity was betwixt 50–70% and photoperiod fixed on 16 hours and 8 h dark. The experiment design composed of 3 nutrient solutions (control non-salt, 40 and 80 mM NaCl) and 4 H_2O_2 treatments (without application control, 50, 100 and 200 mM H_2O_2). 10 plants for one unit of experience were used for each dose of NaCl and each concentration of H_2O_2.The experimental design was set out in a randomized complete block. Globally, 3 replicates per treatment came to 36 experimental unit plots composed by 10 plants each (n = 360 plants). Before sowing, the tomato seeds (cv. Rio Grande) were soaked for 2 hours in varying doses of H_2O_2 solution. One week after seeding, seedlings were transferred into 1L plastic pots planted with unsterile substrate (soil: peat: sand (1/2: 1/4: 1/4)).

Each 3 days, plants were watered with 500 mL of water or with a solution of NaCl (40 and 80 mM).

Studied parameters

Agronomic parameters: At the final harvest, which occurred 21 days after transplanting, the fresh yield of tomatoes was evaluated in eight plants per plot. The shoot biomass was dried in a pressured-air oven at 80°C for three days to determine the shoot's dry biomass. To assess the vegetative growth of the tomato plants, the number of flowers on ten plants for each treatment and block was measured.

Physiological parameter: Membrane integrity was estimated by measuring the electrical conductivity using a digital conductivity meter. The loss of electrolytes was measured at the leaf level and it provides information on membrane integrity. Electrolyte loss was determined using the following method: 5 mm sheet slices obtained from 200 mg of fresh material are placed in tubes containing 20 ml of distilled water. These tubes are incubated at 4°C for 10 hours. We thus measure the electrical conductivity of this water, (EC1). The samples are then autoclaved at 120°C for 15 min, and then cooled to 25°C. The conductivity of the solution of the destroyed leaves, ie (EC2), is again measured.The percentage of membrane damage (Silva *et al*.,2022) was determined as following:

$$EL= (EC1/EC2)*100$$

Measurements were performed on 4 replicates per treatment.

Biochemical parameter: A revelation on gel of the catalase activity (CAT) was carried out on the extracts of total proteins of tomato leaves treated with different doses of Hydrogen peroxide compared to the control (Bradford, 1976).

Revelation on gel of catalase activity (CAT): The method of non-denaturing gels of polyacrylamides was prepared without sodium dodecyl sulfate (SDS). Gels are formed by a separation gel and a compression gel. The first consists of a 30% acrylamide solution of 2% bis-acrylamide, tris-HCl (pH = 8.7), H_2O, persulfate of ammonium (APS) 10% and Tetra methyl-ethylene-diamine (TEMED). Even composition for the second gel with differences in concentration and pH for the trisHCl solution that has a pH of 6.8. After polymerization of the gels, soluble proteins (50µg/sample) are loaded in the wells. The migration on gel is carried out at 70 mA and at 4°C. Finally, the step of revelation consists of incubating the gel in the dark for 30 min with shaking in a solution which contains: potassium phosphate, H_2O_2, EDTA and nitrobluetetrazolium (NBT). After rinsing with distilled water, the gel is transferred to another solution that contains potassium phosphate, H_2O_2, EDTA, acid ascorbic acid and H_2O_2. At the end the gel is incubated in the light without shaking until the appearance of catalase isomorphs (CAT) as a light band on a dark background gel (Pezzoni *et al*., 2018).

Statistical analysis

The statistical analyses were used to compare means of different treatments, we carried out analyzes of variance and comparisons of means by the Duncan test at the 5% probability threshold using the SAS software.

Results

Germination rate: The seed germination started on the 3rd day, as well as final germination rate was 100% under the control condition (Fig. 1). Results showed no change in growth rate when 40 mMNaCl was used compared to conditions without salt. High salt concentration negatively affects tomato growth, and salt stress at 120 mMNaCl can significantly abolish this symptom. Only some seeds germinated on the 5th day, and the final germination rate declined to 40% (Fig. 2). Priming with 50 and 100 mM H_2O_2 doses enhanced the percentage germination under salt stress. In fact, treatment with 100 mM H_2O_2 appeared the most promising application in accordance with the data arranged in Figure 2. However, this positive effect of H_2O_2 on germination was limited and germination rate considerably decreased with the increased concentration of H_2O_2 (from 200 mM).

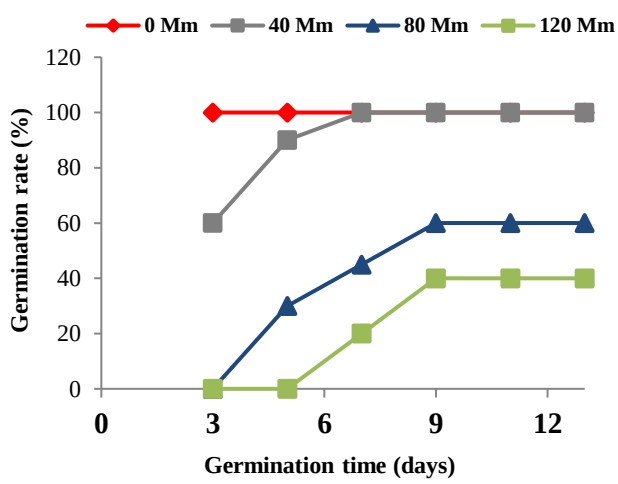

Fig. 1. Effect of NaCl salinity on germination rate of tomato seeds cv. Rio Grande.

Agronomic Parameters

Stems length: The effect of salt stress on the stem length of tomato is shown in Figures 3 and 4. Means comparison for this parameter present a highly significant effect of salt. Indeed, stem length was reduced by 70% at 120 mMNaCl (Fig. 3) in comparison with unstressed treatment which means that the salt inhibited the growth of the tomato plants. For the percentage germination, seed priming treatment with 50 mM H_2O_2 (compared to NaCl treatment) reduced salt stress and progressed the seedling overall performance (Fig. 4). As well as different effects of priming were revealed. The height of the stem is optimal for handling 50 mM H_2O_2 (9 cm) at 40 mMNaCl. However, application of 100 and 200 mM H_2O_2 contributed a decrease of stem length compared to control.

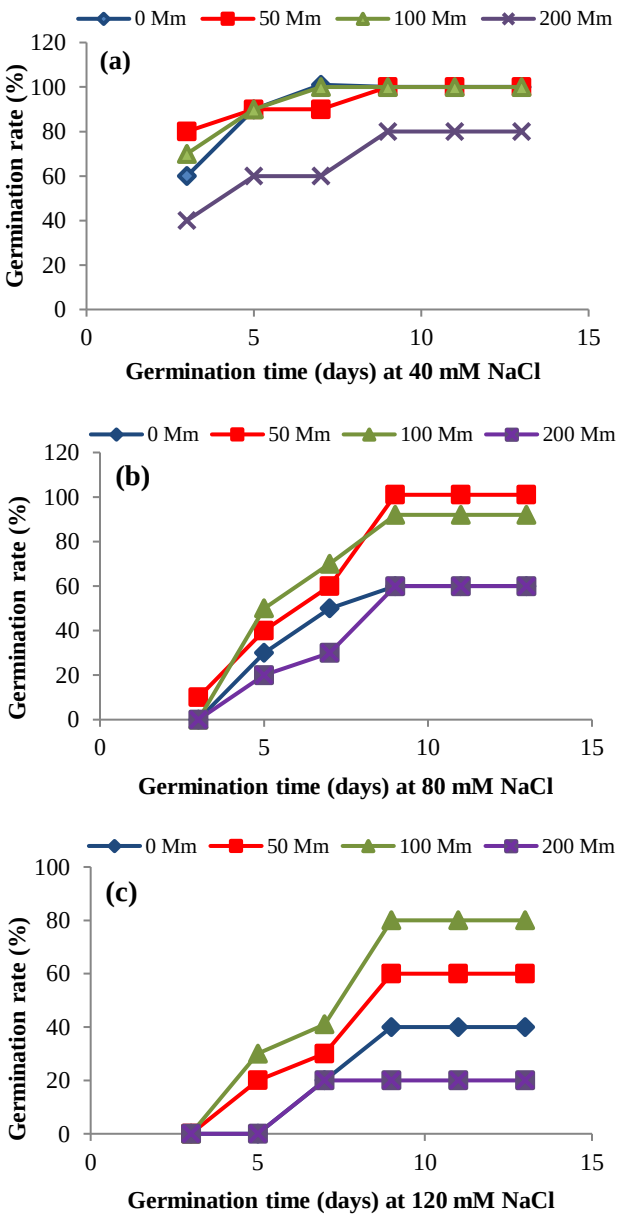

Fig. 2. Effect of different doses of H_2O_2 on germination rate of tomato seeds cv. Rio Grande at different concentrations of NaCl: a: 40 mM NaCl; b: 80 mM NaCl; c: 120 mM NaCl.

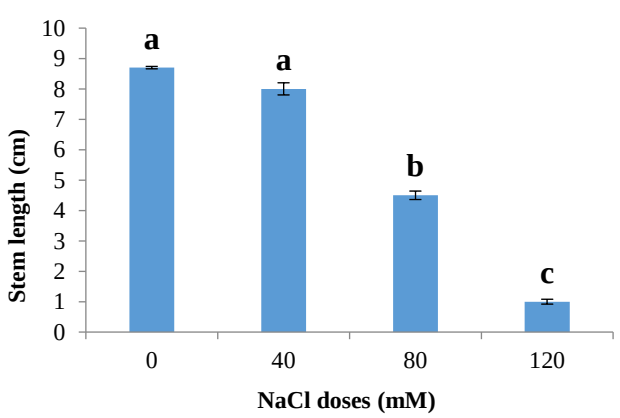

Fig. 3. Effect of different NaCl concentrations on stem length *In vitro* c.v Rio Grande tomato plants.
Data are means SD of four replications, significant means at p≤0.001.

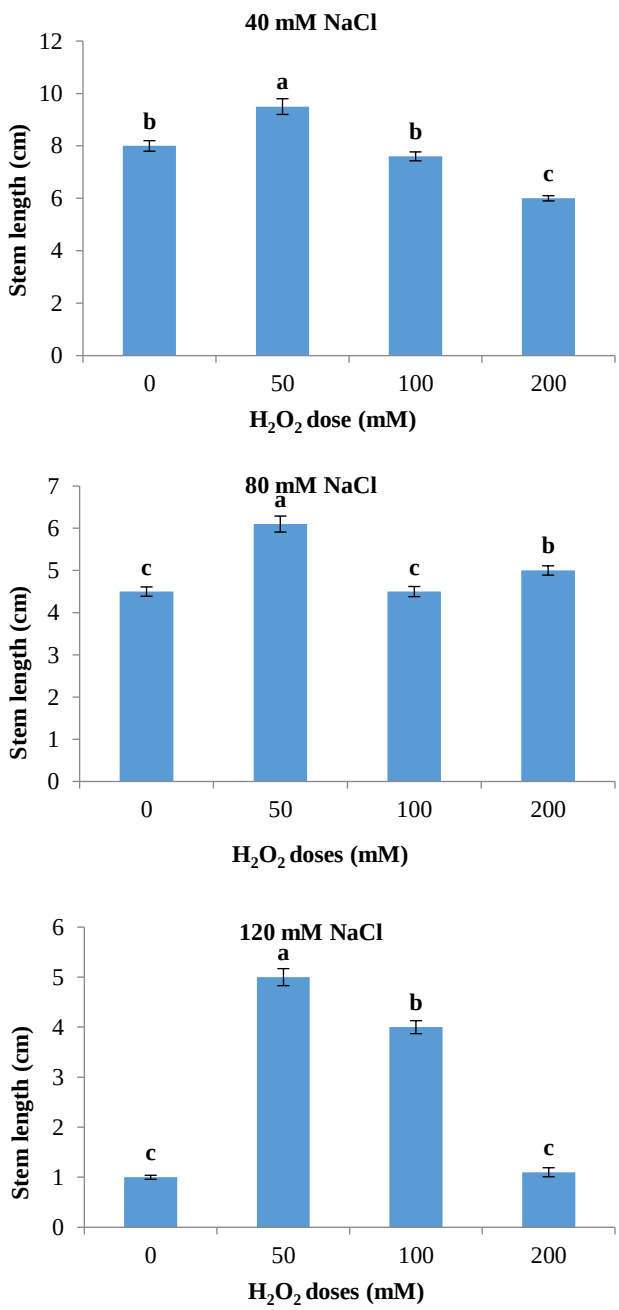

Fig. 4. Effect of four H_2O_2 concentrations applied in pretreatment of salt stress seeds (0,40, 80 and 120 mM) on stem length tomato.
Data are means SD of four replications, significant means at p≤0.001.

Plant growth: A significant (p≤0.05) effect of seed priming on the fresh and dry weight of shoots grown under salt stress has been presented in Table 1. In fact, shoot fresh weight was reduced by 30.8% and 67.04% compared with the control for 40 and 80 mMNaCl. Salt treatment resulted in a decrease in shoot dry weight. This trend was more pronounced with increasing NaCl concentration. Plants salinated with 80 mMNaCl had 8.8% lower shoot dry weight than non-salinated plants (Table 1). Seed priming with H_2O_2 significantly increased shoot fresh weight in both unstressed and salt-stressed conditions. At 40 mM NaCl, priming with

200 mM H_2O_2 clearly increased shoot fresh weight by 17.44% compared with untreated plants. At 80 mMNaCl, treatment with 50 mM H_2O_2 significantly increased fresh shoot weight by 47.13% compared with untreated plants. Whereas seed priming with 200 mM H_2O_2 had no effect on shoot fresh weight compared with untreated plants at this NaCl concentration (80 mMNaCl). At 40 mMNaCl, seed priming with 100 mM H_2O_2 increased shoot dry weight in against the untreated samples. With this concentration, shoot dry weight was improved by 3.86% compared to the untreated plants. Seed priming with 200 mM H_2O_2, however, did not enhance plant dry weight. This parameter was dramatically decreased after 200 mM H_2O_2 application (58.27%).

A significant ($p \leq 0.05$) effect of different amount of Salinity on flower number. Salinity levels are to flower number. At higher salinity level, this parameter was reduced by 71.43% against control. Seed priming with 50 mM H_2O_2 raised the number of flowers by 27.26% at level 40 mMNaCl against non-primed treatment. At higher H_2O_2 level (200 mM H_2O_2), no positive effect was shown by seed priming under salinity.

Membrane integrity: Membrane integrity measurements can provide information on electrolyte loss in tomato leaf cells under salt stress, and on the effect of hydrogen peroxide in salt stress tolerance (Fig. 5). Electrolyte leakage (EL) was improved in tandem with salinity amount rising in comparison with control tomato plants. For untreated plants with H_2O_2, EL was about 30% more than in comparison to non-salinized plants. Treatments with 100 mM H_2O_2 had minimal negative impact on electrophoresis less than 120 mM NaCl amendment. Moreover, a dose of 100 mM H_2O_2diminished 50% of damage compared to the control plants in 80 mMNaCl amendment plants. During this test, the highest leakage rates were reported in control samples tracked by 200 mMH_2O_2-primed plants stressed by 120 mMNaCl.

Catalase activity gel revealed: Figure 6 illustrated catalase activity in tomato leaves in response to seed treatment with hydrogen peroxide. Data analysis showed that seed priming strategies were accompanied by an enhancement of CAT activity. Each band corresponds to one or more isomorphs associated with four tetrameric structure views of enzyme. Our results of CAT activity revelation on gel showed that the activity level of this enzyme varied as a function of the H_2O_2 applied doses.

Table 1. Effects of hydrogen peroxide (H_2O_2) treatment on growth characteristics of tomato c.v Rio Grande under salt stress.

Parameters	NaCl (mM)	Priming treatment with H_2O_2				Statistic effect
		Untreated	50 mM H_2O_2	100 mM H_2O_2	200 mM H_2O_2	
Freshweight	0	108.65 a	95.73 b	80.14 c	70 d	***
	40	74.65 c	75 b	73.13 d	90.86 a	***
	80	3.,9 c	67.9 a	53.4 b	35.5 c	***
Dry weight	0	25 a	18,.10 b	17.35 b	13 c	***
	40	15.96 a	11.6 b	16.6 a	9.3 c	***
	80	22.8 a	16 c	16 c	18 b	***
Flowernumber	0	14 a	12 b	11 b	9 c	***
	40	8 b	11a	9 b	8 b	***
	80	4 b	6a	5 b	4 b	***

Data represent means ± SD, n = 10. Different letters mean significant difference according to a Duncan's multiple range test (***$p \leq 0.001$)

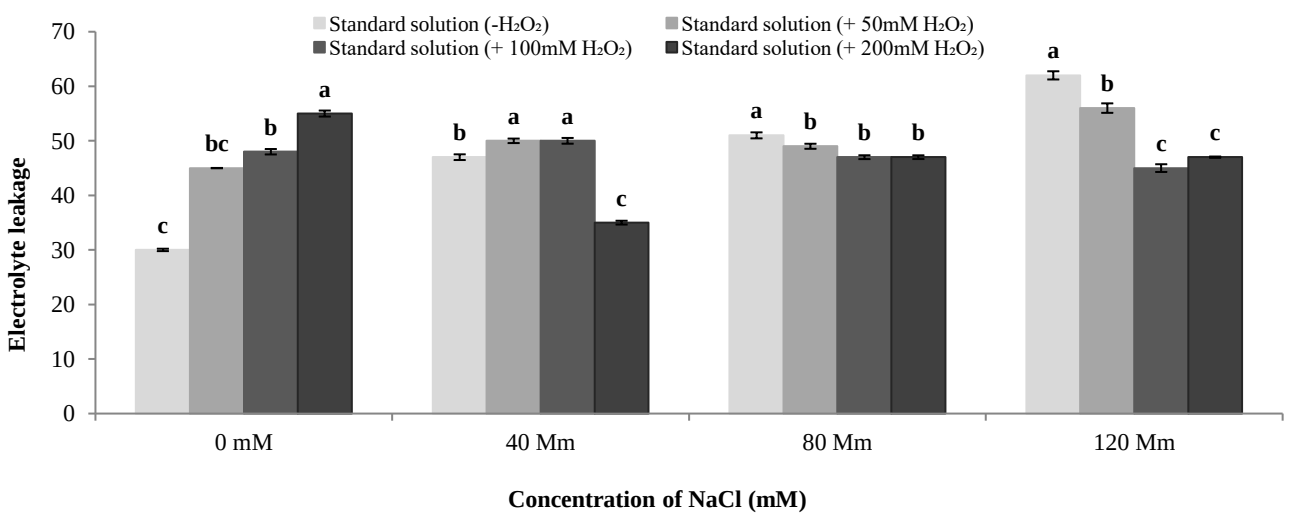

Fig. 5. Effects of four concentrations of H_2O_2 pretreatment of cv. Rio Grande tomato seeds on membrane integrity under increasing salt stress conditions (0,40, 80, 120 mMNaCl).Data are means SD of four replications, significant means at $p \leq 0.001$.

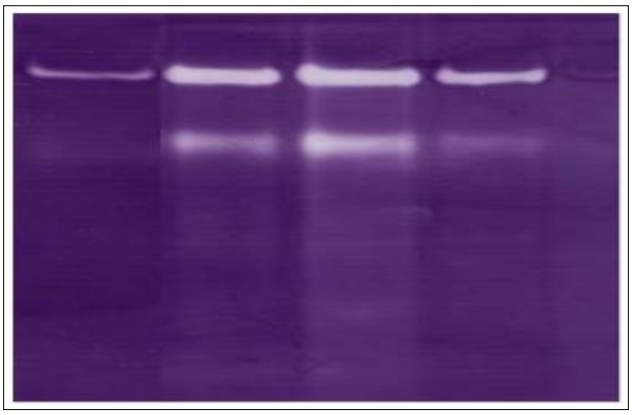

Fig. 6.Non-denaturing gel reveal of catalase activity (CAT) in tomato leaves treated with three H_2O_2 concentrations (0,50, 100, and 200 mM) compared to the control untreated plants.

Discussion

Plant growth: Plants exposed to high salinity can cause significant productivity reductions in the world which cause oxidative damage and ion toxicity (Ilangumaran& Smith, 2017).Therefore, a concerted effort to understand the effects of salinity on plants is critical to addressing the global salinity problem(Chakma *et al.*, 2019). The aim of this study was to test the effectiveness of H_2O_2 priming to induce salt tolerance in tomato plants under salt stress and to identify the main morphological, physiological and biochemical changes involved in this process.

Plant development preliminary stages are a key to anticipate plant development which is highly dependent to seedling germination (Rajabi *et al.*, 2020). This study disclosed that salinity caused a significant abatement in tomato seed germination parameters, suggesting that salt is unfavorable for tomato growth. However, these parameters vary by priming concentration (Lu *et al.*, 2022). It is well known that salinity is negatively correlated with germination (Zhao *et al.*,2021). However, this negative correlation varied with salt concentration. At low NaCl concentrations, it induced seed dormancy whereas at high concentrations it inhibited germination due to Na^+ and Cl^- accretion (Zhao *et al.*,2021). Our results did not revealed any impact of salt on GP at the lowest salt concentration used (40 mM). In general, salt affects germination by altering water and seed germination due to reduced osmotic pressure for germination, lower water absorption (Fasih *et al.*, 2021). Furthermore, high concentrations of Na^+ ions in the medium can lead to osmotic stress resulting in decreased water uptake by plant tissues (Polash *et al.*, 2019). In addition, salt could modify intoxication due to ion toxicity. Disruption of enzymatic activity leads to significant changes in plant growth, such as changes in nucleic acid and protein transfer (Peng *et al.*, 2023), disruption of hormone abundance and reduced reserves (Sachdev *et al.*, 2021). Additionally, salt can stimulate phenolic compounds, which can decrease germination rate (Peng *et al.*, 2023). But also other factors can influence seed germination under saline-alkaline conditions like water, temperature and light (Wang *et al.*, 2022). For shoot and root lengths, this parameter decreased with increasing salt concentration. In fact, roots are the first ones that come into direct contact with salinity soil. They soak up the water and deliver it to aerian organs (Wang *et al.*, 2022). Indeed, plant tissues can be severely affected by salinity. The decrease in stem length of tomato seedlings observed in this study with increasing salinity elucidated that salinity reduces the intercellular carbon dioxide concentration, which in turn reduces the rate of photosynthesis through stomatal closure. Under high salt levels, Na^+ can cause lower transport rate of essential ions such as NO_3^- that reduce the N-containing compounds and ultimately inhibit plant growth (Khaleduzzaman *et al.*, 2021). In fact, H_2O_2 has a positive effect on the response of plants to different types of stress (Danial& Basset, 2024). The results of this study showed that plants treated with H_2O_2 showed a higher germination rate compared to plants treated with NaCl. The optimal concentration of H_2O_2 for germination was 100 mM and 50 mM for Stem length. Seed priming with H_2O_2 significantly increased germination in comparison with the control which is confirmed by results of several reports (Migahid *et al.*, 2019; Marthandan *et al.*, 2020). They have revealed a positive effect of priming on plant growth parameters. Early initiation of transplantation and the maintenance of decomposition and accumulation may account for the rapid germination of the first seeds. The reasons for the above-mentioned effects of seed preparation treatments may be different. Since H_2O_2 is considered as one of the signaling molecules, they may reprogram gene expression (Danial& Basset, 2024), resulting in de novo protein synthesis resulting in a membrane repair mechanism and more storage proteins and other substrates to enhance and synchronize germination. In plants, H_2O_2 is widely used in various biochemical and physiological activities. Ergo its long lifespan and flexibility, H_2O_2 can cross cell membranes and can act as a signaling molecule in stress signaling pathways. These processes can produce different stress-related responses. This research showed that seed germination with an optimal amount of H_2O_2 can enhance plant growth under salinity conditions. H_2O_2 as the crucial agent revealed a rising in dry weight of cannabis crop (Islam *et al*., 2022). Furthermore, seeds grown with H_2O_2 exhibited exorbitant bigger dry weight and root length than wheat under drought conditions (Habib *et al.*, 2020). It has been suggested that seed germination leaves a "stress memory" that activates stress response mechanisms after germination (Lutts *et al.*, 2016).

However, seed germination and increased H_2O_2 decreased the number of sprouts. Despite acting as a signaling molecule, excess H_2O_2 induces oxidative stress, which damages cells and leads to systemic death (Habib *et al*., 2020). Number of flowers was also bettered by H_2O_2 priming. Priming with 50 mM H_2O_2 was the most effective for perfecting number of flowers. It has been conjectured that seed priming may trigger stress responsive set up corresponding to antioxidant stimulation, abscisic acid signaling, and hormonal modulation (Rhaman *et al.*, 2020). It has been suggested that seed priming system can engender physiological acclimatization, that lead to its rise crop yield under stress situation that lead to its increase (Marthandan *et al.*, 2020). In fact, similar results obtained for wheat yield which was raised under drought condition after priming with suitable amount H_2O_2 (Marthandan *et al.*, 2020). Appropriate amount of H_2O_2 can induce stress tolerance by regulating multiple physiological processes such as photosynthesis, stomatal movement, uptake adaptation, and ROS detoxification (Habib *et al.*, 2020).

Membrane integrity: Salt tolerance depended to membrane integrity mechanism. The plasma membrane is a major site of ion-specific salt damage (Sayed&Gadallah, 2019). For that, EL of the plasma membrane is the way to identify halotolerant plants. The results obtained for this parameter prove that the values of electrolyte leakage are directly proportional to increasing salt content. Likewise, the indices of lettuce leaves and roots gradually increased with rising NaCl concentration (Kazamel *et al.,* 2024). Some authors have exhibited this finding in cucumber (Soufi *et al.*, 2023), and strawberry (Wang *et al.,* 2019). In our study, the results of priming seeds with H_2O_2 reduced ion leakage in salinity-stressed tomato plants. This may be due to the stimulation of plasma membrane activity under salt stress by Hydrogen peroxide (Forghani *et al.,* 2021). In the same context, spraying sunflower leaves with H_2O_2 stimulates salt tolerance. On the other hand, in untreated plants, an overproduction of free energy with an excessive secretion of ROS contributes to an aggravated electrolyte leakage (Lephatsi *et al.*, 2021). On the other hand, treatment of plants with H_2O_2 reduced the negative effects of NaCl in a similar way to control plants. The immune system manages so-called antioxidant enzymes with the capacity to trap ROS, such as catalase (CAT). CAT is involved in the cleavage of H_2O_2 into water and oxygen. In fact, the increase in antioxidant activity by H_2O_2 priming is reflected by the stimulation of the expression of various antioxidant enzyme genes, such as superoxide dismutase, catalase and others (Lephatsi *et al.*, 2021; Savvides *et al.*, 2016). Indeed, overexpression of these enzymes can correct the negative effect of salt stress by helping to maintain redox homeostasis. Citing Example, catalase activity is significantly increased in leaves and roots in response to H_2O_2 priming, playing a key role in ROS detoxification (Kesawat *et al.,* 2023).

Conclusion

In conclusion, priming seeds with H_2O_2 contributed to ameliorate germination and growth traits of tomato under salt stress. 100 mM of H_2O_2, used as the initiator, was the most effective concentration as indicated by percentage germination and plant biomass. Seed priming was accompanied by an improvement in activity and a decrease in the amount of ion leakage. Antioxidant signaling appears to be crucial targets of H_2O_2 that lead to increased ion osmotic stress tolerance. It appears that an appropriate concentration of H_2O_2 can be used to mitigate the bad effects of salinity without influencing plants processes to grow under conditions without stress. Limits in NaCl tolerance following H_2O_2 treatment were observed during our trials. And at high concentrations, H_2O_2 can amplify the harmful effects of NaCl. Further clarify the role of H_2O_2 in salt stress tolerance by increasing the duration of soaking using the concentrations that gave the best results. Understand more about the relationship between hydrogen peroxide and the CAT enzyme. And exploit it as an early selection criterion in improvement and creation new varieties tolerant to salt stress.

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