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The Effects of Some Priming Applications on Seed Quality Parameters in Melon (*Cucumis melo* L.) Seeds Under Different Doses of NaCl Stress

Tolga SARIYER[1*], Çağlar KAYA[2]

[1] Çanakkale Onsekiz Mart University, Faculty of Agriculture, Department of Horticulture, Türkiye
[2] Çanakkale Onsekiz Mart University, Faculty of Agriculture, Department of Horticulture, Türkiye
Tolga SARIYER ORCID No: 0000-0002-1844-2996
Çağlar KAYA ORCID No: 0000-0002-7054-3081

**Corresponding author: tolgasariyer@comu.edu.tr*

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Abstract: Salinity of irrigation water and soil-based salinity is an important problem. Priming is an application that increases the germination rate by keeping the seeds in distilled water or different aqueous solutions for a certain period of time. Ipsala variety melon (*Cucumis melo* L.) seeds obtained from Manisa Kırkağaç region were used in the study to determine the effects of different NaCl stress and priming applications on seed quality. Melon seeds subjected to different priming applications were germinated in pure water (0 mM NaCl) and salt stress (140 mM NaCl) conditions. In the study, herbal origin thyme (*Origanum vulgare* L. subsp. *hirtum*), sage (*Salvia officinalis* L.) applications as well as KNO_3 (50 mM) and pure water applications were included as priming applications. In the study, it was determined that increase in the germination rate (%) and germination time (day) parameters which are important quality parameters in seeds, were more pronounced with priming applications. Priming with using sage and thyme plants were more effective respectively in increasing seed quality parameters.

Farklı Dozlarda NaCl Stresi Uygulanan Kavun (*Cucumis melo* L.) Tohumlarında Bazı Priming Uygulamalarının Tohum Kalite Parametrelerine Etkileri

Anahtar kelimeler
Priming,
Kavun,
NaCl stresi,
Çimlenme oranı,
Çimlenme hızı

Öz: Sulama suları ve toprak kaynaklı tuzluluk, önemli bir sorun olarak karşımıza çıkmaktadır. Priming tohumların belli sürede saf su veya farklı sulu solüsyonlarda bekletilmesi şeklinde uygulanan ve çimlenme gücünü artıran bir uygulamadır. Farklı NaCl stresi ve priming uygulamalarının tohum kalitesine etkilerinin belirlenmesi amacı ile yapılan çalışmada Manisa Kırkağaç bölgesinden elde edilen İpsala çeşidi kavun (*Cucumis melo* L.) tohumları kullanılmıştır. Kavun tohumları farklı priming uygulamalarına tabi tutulmuş ardından, saf su (0 mM NaCl) ve tuz stresi (140 mM NaCl) koşullarında çimlendirilmiştir. Çalışmada priming uygulamaları olarak bitkisel kaynaklı kekik (*Origanum vulgare* L. subsp. *hirtum*), adaçayı (*Salvia officinalis* L.) uygulamalarının yanısıra KNO_3 (50 mM) ve saf su uygulamaları yer almıştır. Çalışmada, priming uygulamaları ile tohumda önemli kalite parametrelerinden olan çimlenme oranı (%) ve çimlenme hızı (gün) parametrelerinin artışı daha belirgin olmuştur. Çalışma değerlendirildiğinde, sırasıyla adaçayı ve kekik bitkisi kullanılarak yapılan priming uygulamaları, tohum kalite parametrelerini arttırmada daha etkili olmuştur.

1. INTRODUCTION

The salinity rate in our water resources is increasing day by day. Although there are salts in all surface and underground waters, this salt is transmitted to the soil by irrigation. Salinity affects the water uptake of plants negatively by increasing the osmotic pressure of soil water. Salt accumulation in the soil also negatively affects the physical and chemical properties of the soil [3]. Salt stress causes ion stress in plants as well as osmotic stress. Salt stress has a negative effect on plant growth as a result of the accumulation of ions such as Na^- and Cl^-. The effect of salt stress varies depending on the type of salt, the level of stress, the type and genetic structure of the plant subjected to stress and the developmental stage of the plant [2].

Seeds germinate more easily in laboratory conditions than in farm conditions (farm or nursery). Because they are not exposed to the stress factors (salinity, drought, etc.) in laboratory conditions that they are exposed in farm while germinating. Therefore, it is inevitable that the germination rate will decrease in farm conditions compared to laboratory. This necessitates studies that expose seeds to various stress applications and include treatments that aimed at improving germination in stress conditions.

Various abiotic stresses lead to overproduction of reactive oxygen species (ROS), which are highly reactive and toxic in plants. These reactive oxygen species damage proteins, lipids, carbohydrates and DNA, resulting in oxidative stress [6]. According to the results of a different study [12] on the subject, reactive oxygen species increased in the cells of the elongation zone of the root tip of Arabidopsis with 200 mM NaCl application. Priming is an application that involves leaving the seeds in water under controlled conditions (imbibition) and drying the seed to its initial moisture content with the aim of improving the germination of seeds and initiating early events in germination. Primed seed is brought to a stage where metabolic processes begin with priming before planting. Therefore, primed seed is placed in a more advantageous position than unprimed seed. Primed seed can complete the remaining stages of the germination process (stage three-reimbibition of primed seed) more quickly than untreated seed [22].

Organic priming applications can be done such as using medicinal plants and can be an alternative priming method especially for producers and farmers who engaged in organic farming. Roby et al. [15], stated in their study that thyme (*Thymus vulgaris* L.), sage (*Salvia officinalis* L.), marjoram (Origanum majorana L.) plants have important antioxidant activities and they have a field of use as natural antioxidants. In the study [7], *Origanum vulgare* L. subsp. *hirtum* appears to have antioxidant activity.

Sen et al. [18], determined that NaCl and PEG applications decreased germination rate, increased reactive oxygen species and lipid peroxidation rate in three rice (*Oryza sativa*) cultivars (Neeraja, Vaisakh, Vyttila 6). In their study, they determined that the activities of metabolites as non-enzymatic antioxidant contents as well as antioxidant enzymes were significantly accelerated by different priming techniques. In a study [13], it was reported that priming applications using laurel fruit and moss had better results in pepper seeds while priming applications using dried black thyme and moss had better results in tomato seeds. In the study, it was observed that there was a high increase in germination rate with all priming applications on the third day of germination in tomato.

Özkaynak et al. [14], determined that priming applications had a positive effect on germination in their study using laurel, thyme, seaweed extracts and PEG 6000 in watermelon (*Citrullus lanatus* Thunb. cv. Toraman F1). In the study, it was stated that earlier, more homogeneous and stronger seedlings were obtained with priming applications, and it was stated that laurel, thyme and seaweed extracts could be successfully applied instead of chemical priming.

Rochalska et al. [16], have made priming applications using chamomile (30 and 100% concentration), sage (30 and 100% concentration), chamomile and sage mix (15%+15%) in beetroot (var. Czerwona Kula), sugar beet (var. Janosik) seeds. In the study, they determined that as a result of priming applications, there was an increase in germination ability (%) in both seeds 4 days after sowing, and priming applications had a positive effect on germination rate.

In study [17], with different potassium nitrate applications (0, 0.25, 0.50, 1.00, 1.50, 2.00 %) in rice varieties (KDML105 and RD15), germination percentage values were higher in the priming application with 1% KNO_3 than in the priming application with 2% KNO_3.

Priming applications made by Lara et al. [10], using polyethylene glycol (PEG 6000) -1.1MPa, 50 mM KNO_3, PEG+KNO_3 solutions in tomato. In their study, the priming application using KNO_3 solution had better results than other priming applications in increasing the germination power and germination rate values. The highest nitrate reductase activity was obtained as a result of priming with KNO_3. It was determined that priming with KNO_3 increased the antioxidant system activity, SOD and CAT activities by creating nitric oxide from nitrate reductase.

I another study [19], cumin seeds (*Cuminum cyminum* L.) were germinated in different salt solutions (NaCl, $CaCl_2$, KNO_3), at different concentrations (50, 100, 150 mM) and there was a decrease in the germination percentage under all salt stresses. In study the effect of salt stress was more pronounced with KNO_3 application than $CaCl_2$ and NaCl. In the study, they determined that there was no statistical difference in germination percentage as a result of 50 mM NaCl and $CaCl_2$ applications.

Aydın et al. [1], applied different levels (0, 75, 150, 250 mM) of salt stress (NaCl) in wheat (*Triticum aestivum* cv. Bezostaya), tomato (*Lycopersicon lycopersicum* cv. H55711), beans (*Phaseolus vulgaris* sp. sphearicus) and maize (*Zea mays* cv. Hido) cultivars. Germination rate, germination time, plant salt tolerance index, plant dry weight parameters were negatively affected in all varieties with 150 and 250 mM salt applications. Length of seedlings, root length, fresh weight parameters were negatively affected in all varieties with 75 mM salt application. Among the cultivars included in their study, they determined that the most tolerant cultivar was hybrid corn and the most susceptible cultivar was tomato.

Sivritepe et al. [20], applied priming for 3 days at 20 °C to Hasanbey (*Cucumis melo* L. Hasanbey'') and Kırkagac (*Cucumis melo* L. ''Kırkagac'') melon varieties with using 18 dS m-1 doses of NaCl. In their study, they subjected the seeds to different stress applications by irrigating them with water containing NaCl at doses of 0.3, 4.5, 9.0, 13.5, 18 dS m-1. As a result of their study, when the subjects that were not primed were examined, the germination rate started to decrease at a dose of 4.5 dS m-1 NaCl in Hasanbey variety, while it started to decrease at a dose of 9.0 dS m-1 NaCl in Kırkagac variety. In their study they determined that melon seeds treated with NaCl stress at doses of 4,5, 9 dS m-1 NaCl with priming application and melon seeds treated with dose of 0,3 dS m-1 NaCl were in same statistical group in terms of germination rate. In addition, they determined that the mean germination time increased as the NaCl doses increased. In their studies, germination time decreased with priming application at all NaCl doses except 0.3 dS m-1 NaCl dose. In addition dry weight decreased in melon seedlings with increasing doses of NaCl in unprimed treatments.

The aim of the study is to determine the effects of different NaCl stress and some priming applications (pure water, herbal, chemical origin) on seed quality of melon (*Cucumis melo* L.) obtained from Manisa Kırkağaç region in Turkey where is an intense melon production.

2. MATERIAL AND METHOD

The research was carried out in the laboratory of Çanakkale Onsekiz Mart University, Faculty of Agriculture, Department of Horticulture in 2022. In the study, the seeds of Ipsala variety melon (*Cucumis melo* L.) grown in the Kırkagac region of Manisa province were used. Melon seeds, which were separated from the fruits harvested 50 days after full bloom in 2021, were dried using natural methods without any chemical treatment. The seeds were washed with distilled water for surface sterilization and dipped in 3% sodium hypochlorite solution for 10 seconds to disinfect the seed surface from fungi with the help of a sieve. The experiment was established according to the randomized plot design with 3 replications. 30 seeds were used in each replication in the experiment. The seeds were primed for 24 hours using solutions of dried thyme (*Origanum vulgare* subsp. hirtum), sage (*Salvia officinalis* L.) plants [16], KNO_3 (50 mM) and distilled water. The sage and thyme materials used in the study were obtained from a commercial store in the Çanakkale region. Primed and untreated seeds were germinated in purified water and high salinity (140 mM NaCl) conditions at 25°C. The seeds weighed before the priming application, were dried after the application in a shaded and ventilated environment until they reached their weight before the priming application. Seeds were arranged between 40×40 cm filter papers and watered with distilled water with or without NaCl in equal amounts on the first day and in equal amounts on the 7th day so that no dry areas were left on the filter papers. Seeds were kept in dark conditions in plastic bags for 14 days.

In the priming application using thyme, 5 g of dry thyme was put into 500 ml of water. The mixture was boiled and allowed to cool. 25 ml of the solution was taken, 175 ml of water was added and the application was made [13], [14]. The same application in [13], [14] was made using 5 g of sage. Sage *(Salviae folium)* was used in priming application by [16] as primed for 24 hours in red beet and sugar beet. Study consist of control, priming using 0 mM NaCl+dry thyme, priming using 0 mM NaCl+dry sage, priming using 0 mM NaCl+KNO_3, priming using 0 mM NaCl+pure water, priming using 140 mM NaCl, 140 mM NaCl+dry thyme, priming using 140 mM NaCl+dry sage, priming using 140 mM NaCl+KNO_3, priming using 140 mM NaCl+pure water applications. Thus, the study consisted of 10 subjects.

Analysis and measurements included in the study:

Germination Rate (%): Seeds that rooted out and reached 2 mm in length were stated as germinated. Seed lots were counted at the same time each day. Germination rate determined as = Number of germinated seeds Total number of seeds^{-1}.

Mean germination time (days): It was determined using the= $\sum \frac{n*D}{\sum n}$ formula [5].

n: Number of seeds germinated in D day hour^{-1}
D: Number of days from the start of germination
Σn: Total number of germinated seeds

Hypocotyl Length (mm): It was determined by measuring the length from the cotyledon leaves to the radicle thickness with the help of a caliper with a sensitivity of 0.01.

Radicle Length (mm): It was determined by measuring the length from the tip of the radicle to the end of the radicle with the help of a caliper with a sensitivity of 0.01. Radicular Collar Thickness (mm): It was determined by measuring the thickness of the radicula just before the hypocotyl with the help of a caliper with a precision of 0.01.

Seed Vigor Index: [21].

SAS.8.0. programme was used in making statistical analyzes in the trial. Analysis of variance was performed, and the LSD (P<0.05) test was used to compare the differences between the means of the data.

3. RESULTS AND DISCUSSION

Table 1. Effects of Different NaCl and Priming Applications on Germination Rate (%)

	Control[b]	Thyme[b]	Sage[b]	KNO_3[b]	Pure Water[b]
Control[a]	91.11 ^{A}	95.55 ^{A}	94.44 ^{A}	93.33 ^{A}	92.22 ^{A}
140 mM NaCl[a]	61.11 ^{D}	74.44 ^{C}	85.55 ^{B}	71.11 ^{C}	73.33 ^{C}

LSD P<0.05= 4.5247[c]
[a]: NaCl levels, [b]: Priming applications, [c]: LSD (P < 0.05) value

When the germination rate values were examined, it was seen that the germination rate values did not differ statistically ($P < 0.05$) as a result of priming applications in the subjects that did not apply NaCl stress. When the unprimed subjects were examined, it was determined that the germination rate value decreased significantly ($P < 0.05$) as a result of NaCl application. It was concluded that priming application using sage plant had the highest value among the subjects to which NaCl was applied (Table 1). In a study [12], reactive oxygen species that cause oxidative stress increased with NaCl stress. In another study [18], it was determined that non-enzymatic antioxidant content and activities of antioxidant enzymes were accelerated by priming techniques. In a study, it was stated that priming applications affect germination positively in watermelon [14]. In another study [19], it was mentioned that the germination percentage of cumin decreased under different salt stresses (NaCl, $CaCl_2$, KNO_3).

Table 2. Effects of Different NaCl and Priming Applications on Germination Time (days)

	Control[b]	Thyme[b]	Sage[b]	KNO_3[b]	Pure Water[b]
Control[a]	5.35 F	5.3 F	5.48 EF	5.33 F	5.37 F
140 mM NaCl[a]	6.76 A	5.94 BC	5.66 DE	6.15 B	5.79 CD

LSD P<0.05= 0.2562[c]
[a]: NaCl levels, [b]: Priming applications, [c]: LSD ($P < 0.05$) value

When the germination time values were examined, it was determined that the germination time increased slightly ($P < 0.05$) compared to control in the priming application using sage in subjects without NaCl stress. When the unprimed subjects were examined, it was observed that the germination time was increased statistically ($P < 0.05$) as a result of the NaCl application compared to the control application. When the subjects with NaCl application were examined, it was observed that the longest germination time was achieved in the subject that was not primed, the fastest germination was achieved in the application of priming using sage and this application was followed by pure water, thyme, and KNO_3 applications, respectively (Table 2). In a study [16], it was determined that priming applications had a positive effect on the germination rate of red beet and sugar beet.

Table 3. Effects of Different NaCl and Priming Applications on Hypocotyl Length (mm)

	Control[b]	Thyme[b]	Sage[b]	KNO_3[b]	Pure Water[b]
Control[a]	92.39 C	113.97 A	107.66 B	91.81 C	89.8 C
140 mM NaCl[a]	22.53 E	28.73 D	26.41 DE	25.3 DE	23.19 E

LSD P<0.05= 4.7614[c]
[a]: NaCl levels, [b]: Priming applications, [c]: LSD ($P < 0.05$) value

When the hypocotyl length values were examined, it was observed that priming applications using thyme and sage had higher values than the control application ($P < 0.05$) and thyme application had the highest value in the subjects without NaCl stress. Hypocotyl length decreased significantly ($P < 0.05$) with 140 mM NaCl application. Hypocotil length of not primed seeds and seeds primed with pure water were in same statistical group ($P < 0.05$) in subjects which were treated with 140 mM NaCl. Priming application using thyme had highest hypocotyl length value in subjects treated with 140 mM NaCl (Table 3). In a study [1], 75 mM NaCl application in wheat, tomato, beans and corn reduces plant seedling and root length. In another study [14], early maturing, more homogeneous and stronger seedlings were obtained with priming applications.

Table 4. Effects of Different NaCl and Priming Applications on Radicle Length (mm)

	Control[b]	Thyme[b]	Sage[b]	KNO_3[b]	Pure Water[b]
Control[a]	98.17 B	114.54 A	112.64 A	99.63 B	115.73 A
140 mM NaCl[a]	36,39 E	48,47 C	40,09 DE	49 C	44.44 DC

LSD P<0.05= 6,0948[c]
[a]: NaCl levels, [b]: Priming applications, [c]: LSD ($P < 0.05$) value

When the radicle length values were examined, it was determined that the values in the priming applications using thyme, sage and distilled water were in the same statistical group ($P < 0.05$) and were higher than the control application in the subjects without NaCl stress. It was observed that radicular length values decreased significantly ($P < 0.05$) with 140 mM NaCl application. When the subjects with 140 mM NaCl stress were examined, it was determined that the radicula length values of the subjects that were primed with thyme and KNO_3 were in the same statistical group ($P < 0.05$) and they had the highest values among the subjects at this NaCl stress level (Table 4). Radicle length decreased with increasing NaCl stress in some halophyte species (*Limonium sinense* Kuntze, *Glycine soja* sieb., *Sorghum sudanense* Stapf.) [11] and rice (*Oryza sativa*) genotypes [8].

Table 5. The Effects of Different NaCl and Priming Applications on Radicular Collar Thickness (mm)

	Control[b]	Thyme[b]	Sage[b]	KNO_3[b]	Pure Water[b]
Control[a]	1.57 EF	1.53 F	1.76 ABCD	1.65 DEF	1.68 CDE
140 mM NaCl[a]	1.85 AB	1.86 AB	1.72 BCD	1.9 A	1.8 ABC

LSD P<0.05= 0.1442c
a: NaCl levels, b: Priming applications, c: LSD ($P < 0.05$) value

When the radicular collar thickness values were examined, fluctuations were observed and it was determined that the radicle thickness values mostly increased with priming applications (Table 5).

Table 6. The Effects of Different NaCl and Priming Applications on Seed Vigor Index

	Control[b]	Thyme[b]	Sage[b]	KNO_3[b]	Pure Water[b]
Control[a]	17358.9 D	21841.2A	20817.5 B	17873.5 D	18946.7 C
140 mM NaCl[a]	3599.1 F	5745.6 E	5688.1 E	5278.2 E	4954.9 E

LSD P<0.05= 980.23
a: NaCl levels, b: Priming applications, c: LSD ($P < 0.05$) value

When the seed Vigor Index values were examined, it was determined that the KNO_3 application and the control application were in the same statistical group ($P < 0.05$)

in the subjects without NaCl stress. The highest seed vigor index value was determined in the priming application using thyme. It was observed that the seed vigor index decreased significantly with NaCl stress. Priming applications were significantly ($P < 0.05$) and equally effective in increasing the seed vigor index in 140 mM NaCl application (Table 6). It has been mentioned in some studies [9], [4], that priming applications in pepper (*Capsicum annuum* L.) and safflower (*Carthamus tinctorius*) are effective in reducing the negative effect of salt stress on the vigor index.

4. CONCLUSION

When the study was evaluated, it was determined that priming applications using thyme, sage, KNO3 and pure water were effective in reducing the negative effects of NaCl stress (140 mM) in all parameters except pure water application on hypocotyl length.

When evaluated statistically, priming applications did not affect the germination rate in subjects without NaCl stress. In subjects without NaCl application, priming applications did not affect positively to the germination time in subjects except sage application. When seed vigor index values were evaluated, thyme application had the highest value in subjects without NaCl stress.

When evaluated statistically, in reducing the negative effects of NaCl stress, sage application was more effective than other priming applications in terms of germination rate and time parameters and thyme application was more effective than other priming applications in terms of hypocotyl length. KNO3 and pure water applications never had the best effect on reducing the negative effects of NaCl stress statistically among priming applications but KNO3 application had better results against pure water application in terms of germination time, radicle length, hypocotyl length, radicular collar thickness applications.

When the data were evaluated, it was seen that sage and thyme applications were the most effective priming applications respectively.

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