

## Assesment of seed germination and seedling growth of *catharanthus roseus* under salinity stress and priming

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### Abstract

An experiment was conducted based on a factorial in a completely randomized design with three replications. The factors include four salinity levels using sodium chloride and five priming levels. The results showed seed priming was effective in modulating the effect of stress at all salinity levels. Potassium nitrate priming was more effective than ascorbic acid. In treatment without salt stress, the application of priming in some levels, increased seedling length and seedling vigor index compared to the control.

**Keywords:** Periwinkle; Potassium nitrate; Salinity, Ascorbic acid

### 1. INTRODUCTION

Salinity stress is one of the abiotic stresses that severely limits the development and growth of plants in green spaces [1]. This stress has a negative effect on the growth and yield of the crop and causes a delay in germination, decrease in germination percentage and seedling growth [2].

The increase in population and the decrease in access to qualified water caused an increase in irrigation of green spaces with saline water, which poses serious problems for the establishment of plants in these spaces [3]. One of the most sensitive periods in the life of plants is the stage of germination and plant establishment in the soil (1). Salinity limits seed germination, growth and establishment of plants [4]. Seed priming has been reported as a useful strategy in modulating the effects of salinity stress in many plants [5].

Seed priming can increase the activity of antioxidant enzymes and raise the tolerance to abiotic stresses such as salinity and drought [6]. In wheat, seed haloprime with KCl and 2 CaCl<sub>2</sub> increased germination, decreased the amount of proline and Na<sup>+</sup> and stimulated antioxidant activities in salinity stress and increased plant resistance to stress [5].

Ascorbic acid is known as a substance to improve the defense system of plants against salt stress [7].

Priming with ascorbic acid improved germination indexes and enzyme activity of radish under drought stress [8]. Also, the positive effect of prime with potassium nitrate was reported in kale (*Brassica oleracea* var. capitata) [9], caper (*spinosa Capparis*) [10], and *Cannabis sativa* [11].

Madagascar periwinkle (*Catharanthus roseus*) belongs to the Apocynaceae family, which is an important ornamental plant in gardens and green spaces, which is cultivated as a pot, border and cover. This plant is sensitive to some environmental stresses such as salinity and drought stress, and in terms of growth and beauty, the plant undergoes a significant decline under stress conditions [12,13] Considering the ornamental value of the Madagascar periwinkle in green spaces and the major propagation of this plant through seeds, the use of saline water in the irrigation of green spaces is due to the lack of quality water and the lack of information on the effect of priming on seed germination. This plant is under salinity stress, this study was conducted with the aim of investigating the effect of priming on the germination of plant seeds under salinity stress conditions.

### 2. MATERIALS AND METHODS

This research was conducted in 1402 in the Physiology Laboratory of Horticultural Sciences and Green Space Engineering, Faculty of Agriculture, Ferdowsi University of Mashhad.

This study was conducted as a factorial experiment based on a completely randomized design with three replications. The first factors included four salinity levels (0, 4, 8 and 12 deci-Siemens/m) and five levels of seed priming (potassium nitrate 100 and 200 mg/l and ascorbic acid 100 and 200 mg/l and distilled water).

After disinfecting the seeds, 25 seeds were placed in each petri dish with a diameter of 9 cm and placed on two Whatman filter papers, salinity for each petri dish were applied. Then, the Petri dishes were closed by parafilm and placed in the germinator with a temperature of 25<sup>o</sup>c and 12 hours light/dark period.

Germinated seeds were counted daily. The criterion for germination was 2 mm of radicle exit from the seed. At the end of the experiment, the length of the root and stem were measured.

The parameter evaluated in this experiment were:

$$\text{Germination percentage} \quad GP = (n/N) * 100 \quad (1)$$

GP: germination percentage, n: number of germinated seeds, N: total number of seeds [14].

$$\text{Germination rate} \quad GS = \sum (ni / Ti) \quad (2)$$

GS: the germination rate, ni: the number of germinated seeds per count, Ti: the number of days until count [15].

Seedling length

$$\text{Seedling vigor index} \quad SVI = GP\% \times SL \text{ (mm)} / 100 \quad (4)$$

SVI: the seedling vigor index, GP: the percentage of germination, SL: the average seedling length [16].

The data was analyzed by SAS software version 9.4 and comparison of means was done through LSD test at the five percent probability level.

### 3. RESULTS

The results of analysis of variance showed that salinity stress had a significant effect on all investigated traits, including germination percentage, germination rate, average daily germination, seedling length index at the probability level of 1% (Table 1).

**Table 1. Analysis of variance for germination and seedling growth characteristics of Madagascar periwinkle under priming and salinity stress**

Mean square					
Sources of variation	df	Germination percentage	Germination rate	Seedling length	Seedling vigor index
(A)	3	9570.40**	67.78**	6294.73**	8722.81**
Salinity stress					
(B)	4	115.73 <sup>ns</sup>	0.19 <sup>ns</sup>	57.81**	98.96**
Priming					
Interaction AB	12	112.18 <sup>ns</sup>	0.45 <sup>ns</sup>	101.57**	134.37**
Error	40	6026.67	23.49	1862.67	2109.19
CV (%)		17.42	16.67	16.04	18.98

\*, \*\* and ns are statistically significant at the probability levels of 5, 1% and not significant, respectively.

## 3.1. Germination percentage and Germination rate

with the increase of salinity, the germination percentage and germination rate of seeds decreases and there was significant difference between salinity levels, the lowest germination percentage and germination rate were observed at 12 des/m salinity level (Fig. 1 A, B).

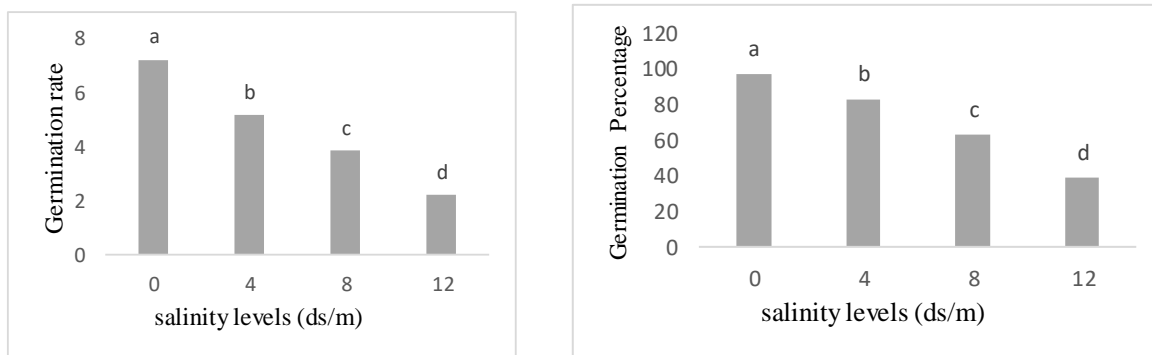


Figure 1. Mean comparison of germination percentage (A) and germination rate (B) in Madagascar periwinkle, similar letters have no significant difference at 5% probability

## 3.2. Seedling vigor index

The interaction effect of salinity stress and priming was significant at the 5% probability level for seedling length index (Table 1). With increasing salinity level, the seedling length index decreased. At each salinity level, a significant difference was observed between different levels of priming (Fig. 2). The highest amount for this trait was observed at 0 des/m salinity stress by application of 100 ppm ascorbic acid and then the level of 100 ppm of potassium nitrate.

At 4, 8 and 12 des/m salinity, the seedling length index was lower at the control level (water priming) compared to the priming with potassium nitrate and ascorbic acid. At the level of 4 des/m, the highest value for seedling length index was observed at 100 ppm of potassium nitrate, which was significantly different from the control level (Fig. 2). At 8 and 12 des/m salinity, application of 200 and then 100 ppm potassium nitrate had the highest amount for the seedling length index compared to other levels (Fig. 2).

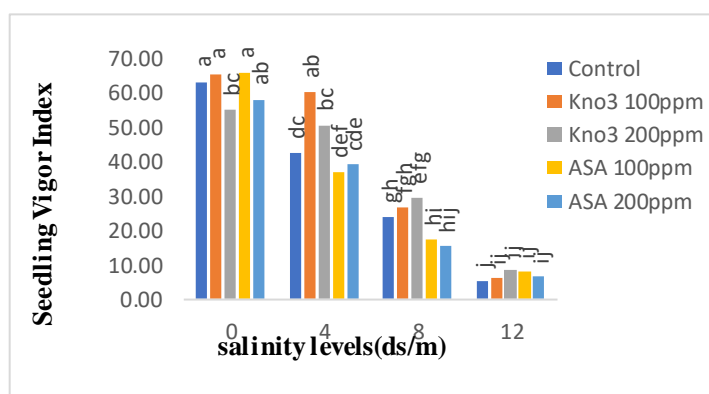


Figure 2. Interaction effect of seed priming and salinity stress on Madagascar periwinkle seedling vigor index, similar letters have no significant difference at 5% probability

## 3.3. Seedling length

Based on the results obtained from the analysis of variance (Table1), it was observed that salinity stress, priming and the interaction effect of salinity and priming on seedling length were significant at 1% probability level (Table 1). With increasing salinity level, seedling length decreased. Application of priming at different salinity levels, led to significant difference for seedling length compared to the control (priming with distilled water).

At 4 des/m salinity, priming with 100 and 200 ppm of potassium nitrate had longer seedling length than the control, and also at 8 des/m salinity, priming with 200 ppm Potassium nitrate was more than the control and had a significant difference with each other (Fig. 3). Among the different treatments, the highest amount of seedling length was observed at 100 ppm ascorbic acid and at the 0 des/m salinity, and the lowest amount was observed at the control and 12 des/m salinity (Fig. 3).

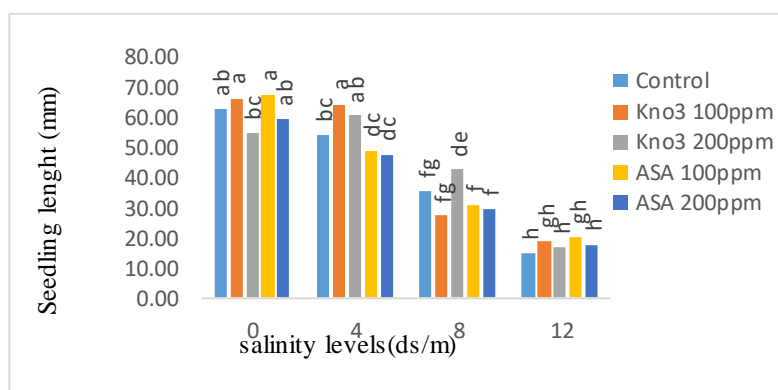


Figure 3. Interaction effect of seed priming and salinity stress on Madagascar periwinkle seedling length, similar letters have no significant difference at 5% probability

## 4. DISCUSSION

By increasing salinity levels, the growth, germination and weight of plants decreases [7, 17, 18], which was similar to the results observed in this experiment. Salinity causes a delay in water absorption and thus affects the germination process [17].

Seedling length reduction due to salinity was also observed in other plants, including mung bean (*Vigna radiata*), soybean (*Glycine max*) [5], and *Cassia fistula* [4]. Plants exposed to salt stress slow down their growth in two stages, in the first stage they consume energy to deal with salt stress and salt solutions, and in the second stage due to the accumulation and toxicity of salt in parts such as chloroplasts and mitochondria. It causes a decrease in growth and development [19].

Application of potassium nitrate and ascorbic acid at all salinity levels, had positive effect on the growth of seedlings, and the seeds primed with these compounds had better growth, and in the meantime, priming with potassium nitrate was more effective than ascorbic acid. The effective use of potassium nitrate on germination components has also been reported by Ahmadvand et al [1].

In general, based on the results obtained from this experiment, it was found that different levels of priming with potassium nitrate and ascorbic acid had a positive effect on the seedling vigor index.

The positive effects of priming can be due to the increased activity of enzymes in the seedling axis, which increases the transfer of compounds such as proteins, amino acids to the seedling axis [1].

## 5. CONCLUSION

The results of this research showed that priming improved seedling vigor index and seedling length. The application of priming had no effect on the components of the germination percentage and germination rate, and it seems that the application of priming had an effect on the components after seed germination and on the growth parameters. Among the priming, it seems that the use of potassium nitrate is more effective compared to ascorbic acid.



## 6. ACKNOWLEDGMENT

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