

Effect of Phasic Pretreatment of Sodium Chloride on Seedling Growth of *Vigna Radiata* (L.)

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ABSTRACT: To study the effect of phasic pretreatment of Sodium Chloride (NaCl) on seedling growth of *V. radiata* L. the whole imbibition period was divided into six equal phases of 2 hours each. Maximum inhibition was observed in the mid phase treated seedlings. Cytotoxic effects on cell divisions were also noticed in the treated sets.

Keywords: *V. radiata* (L.), Sodium Chloride (NaCl), Enzymes, Phasic pretreatment.

I. INTRODUCTION:

Salinity is a major threat to modern agriculture causing inhibition and impairment of crop growth and development. Salinizations are on the increase mainly due to poor irrigation drainage or agricultural practices (Saha P., et.al., 2010). The negative effects of salinity, owing to increase in Na^+ and Cl^- ions, disturb the homeostasis of essential nutrients (Ghogdi E. A., et.al., 2012 and Roychoudhury A., et.al., 2013), leading to both hyperionic and hyperosmotic stress. The effects may be membrane damage, enzymatic inhibition, alterations in levels of growth regulators, nutrient imbalance, reactive oxygen species (ROS) generation leading to DNA damage and cell death (Gill S. S., et.al., 2010 and Sharma P., et.al., 2012). Typical sodium toxicity symptoms are leaf burn and dead tissue along the outside edges of leaves. In contrast, the symptoms of chloride toxicity occur initially at the extreme leaf tip. The chloride ion can be taken up by plant roots and accumulate in the leaves. Excessive accumulation may cause burning of the leaf tips or margins, bronzing and premature yellowing of the leaves.

Pulses are the best dietary source of proteins and they play a very important role to fulfill the requirements of rapidly increasing population. Mung bean (*Vigna radiata* L., Wilczek) is synonymous with *Phaseolus aureus*

Roxb. The mung bean is a legume (Family: Fabaceae) cultivated for its edible seeds and sprouts across Asia. They are warm season annuals, highly branched and having trifoliolate leaves. Both upright and vine types of growth habit occur in mung bean, with plants varying from one to five feet in length. The pale yellow flowers are borne in clusters of 12–15 near the top of the plant. Mature pods are variable in color (yellowish-brown to black), about five inches long, and contain 10 to 15 seeds. They can be eaten whole or made into flour, soups, porridge, snacks, bread, noodles and ice-cream. Mung sprouts are high in protein (21%–28%), calcium, phosphorus and certain vitamins.

The present work was undertaken to screen the impact of sodium chloride on seedling growth of *V. radiata* L. The purpose of the study was to know whether the entire period of imbibition or a particular phase of imbibition is important for maximum inhibition of seedling growth.

II. MATERIAL AND METHODS :

Mung (*V. radiata* cv. Hum-16) seeds were used for experiments. Seeds of test plants were selected on the basis of uniformity in size, shape, colour & weight. Seeds were surface sterilized with 0.1% HgCl_2 solution and thoroughly washed with distilled water. The different concentrations of sodium chloride (10mM and 100 mM) were prepared in double distilled water. For phasic pretreatment of Sodium Chloride (NaCl), the whole imbibition period was divided into six equal phases of 2 hours each. Fig.-1 shows the six phasic pretreatment regimes (regimes-1, 2, 3, 4, 5 & 6) and a control set of *V. radiata*. In the control set, seeds were imbibed in distilled water for their whole imbibition period, while on the other hand for phasic pretreatment, seeds were treated with 10mM and 100mM of sodium chloride solutions separately.

Fig.-1: Phasic pretreatment of Sodium Chloride (NaCl)

Imbibition Period	0-2 hrs.	2-4 hrs.	4-6 hrs.	6-8 hrs.	8-10 hrs.	10-12 hrs.
Control	Water					
Treated Set (1)	NaCl	Water				
Treated Set (2)	Water	NaCl	Water			
Treated Set (3)	Water		NaCl	Water		
Treated Set (4)	Water			NaCl	Water	
Treated Set (5)	Water				NaCl	Water
Treated Set (6)	Water					NaCl

In the treated set-I, seeds were imbibed in sodium chloride solution during the first phase i.e. between 0-2 hrs, followed by imbibition in distilled water for remaining period i.e. 2-12 hrs. In set-2, seeds were imbibed in sodium chloride solution during the second phase i.e. between 2-4 hrs, while in the preceding and following periods in distilled water i.e. 0-2 hrs and 4-12 hrs. In set-3, seeds were imbibed in sodium chloride solution during the third phase i.e. between 4-6 hrs, while the rest of phases i.e. 0-4 hrs and 6-12 hrs. in distilled water. In set-4, seeds were imbibed in sodium chloride solution during the fourth phase i.e. between 6-8 hrs, while the rest of phases i.e. 0-6 hrs and 8-12 hrs. in distilled water. In set-5, seeds were imbibed in sodium chloride solution during the fifth phase i.e. between 8-10 hrs, while the rest of phases i.e. 0-8 hrs and 10-12 hrs. in distilled water. Similarly,

in set-6, seeds were imbibed in sodium chloride solution during the sixth phase i.e. between 10-12 hrs, which was preceded by imbibition in distilled water. Thereafter, the control as well as treated seeds were washed thoroughly with distilled water and transferred to petridishes lined with moist filter paper and kept in dark for seedling growth. The experiment was performed in triplicate. The growth parameters like plumule and radicle length were observed on 10th day after radicle emergence. The data observed in the experiment, were statistically analyzed for the calculation of standard error.

For mitotic studies root tips of germinating seeds were fixed in acetic alcohol (1:3) and squashed in 2% of acetocarmine. Number of normal and abnormal cells and types of chromosomal abnormalities were noted in all concentrations.

III. RESULT AND DISCUSSION:

The Results of phasic pretreatment on seedling growth obtained in the experiments are shown in tables-1 & 2 and fig.- 2 to 5.

Effect of phasic pretreatment of NaCl (10mM and 100mM) on seedling growth of *V. radiata* L.

Table-1 :

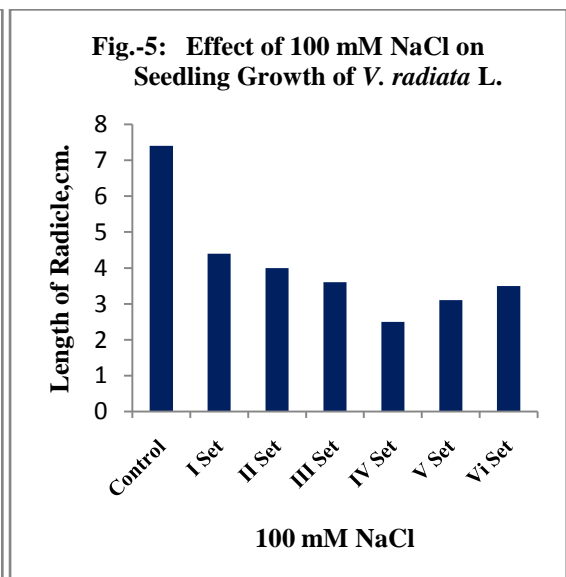
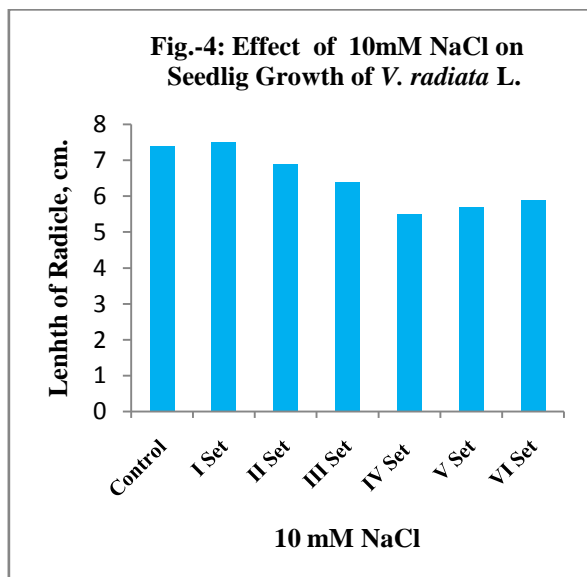
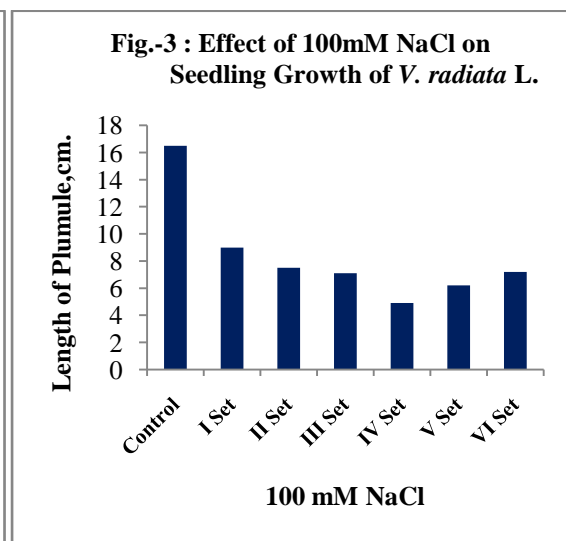
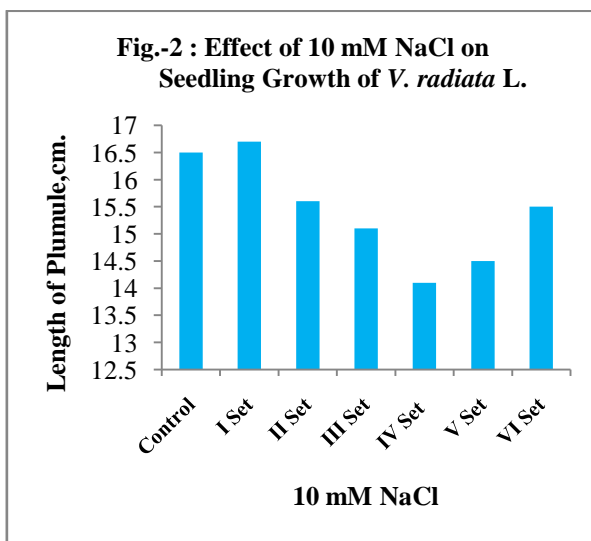
Concentration of NaCl	Length of Plumule after 10 days (cm.)					
	I	II	III	IV	V	VI
Control	16.5 ± .22					
10mM NaCl	17.0 ± .25	15.6 ± .22	15.1 ± .27	14.1 ± .33	14.5 ± .22	15.5 ± .22
100mM NaCl	9.0 ± .24	7.5 ± .22	7.1 ± .25	4.9 ± .22	6.2 ± .25	7.2 ± .33

The average of three triplicates ± S.E

Table-2 :

Concentration of NaCl	Length of Radicle after 10 days (cm.)					
Control	7.4± .33					
Treated Sets	I	II	III	IV	V	VI
10mM NaCl	7.5 ± .23	6.9 ± .22	6.4 ± .33	5.5± .25	5.7 ± .25	5.9 ± .22
100mM NaCl	4.4 ± .24	4.0 ± .25	3.6 ± .22	2.5± .22	3.1 ± .33	3.5 ± .25

The average of three triplicates ± S.E



The phasic pretreatment studies showed that the lower concentration (10mM) of sodium chloride had no significant effect on seedling growth while the higher concentration of sodium chloride was inhibitory to seedling growth.

Noteworthy point was that the extent of inhibition varies in different phases being maximum in mid-phase (regime-4). At higher concentration (100mM), the length of radicle was 33.78% and plumule length was 29.69% of control respectively

in mid-phase treated sets. The mid phase treatment regime was more effective as at this phase there was maximum inhibition of seedling growth was noticed while in the remaining phases the inhibitory effect was in between the initial and mid-phase. From phasic pretreatment studies, it is inferred that sodium chloride uptake by seeds takes place in all phases but is maximum in mid phase. It appears that because of maximum uptake of sodium chloride in this phase, there is maximum inhibition of enzymes. Due to this reason maximum inhibition of seedling growth was noticed in mid phase treated sets. Present observations on seedling growth are comparable with the earlier findings reported by Dubey R.C., et. al.(1987), Prasad, et.al, (1981) and Singh A.,(1996),

The most remarkable response of sodium chloride on cell division is steep decrease in mitotic activity with gradual increase in concentration which was quite normal in control. Lowest MI was noticed in mid- phase treated seedlings at higher concentration. This finding clearly indicates that maximum absorption of sodium chloride during mid phase damage some of the enzyme system involved in the metabolism and repair mechanism, thus directly affects the cell divisions and cell elongation , hence retardation in the growth of seedlings. Similar cytotoxic effects have been reported by Grant W.F., et. al.(1978), Fojtova M.,et.al.(2000) and Smruti G., et.al.(2018).

The present investigation clearly indicates the inhibitory nature of sodium chloride on *V. radiata* L., although very low concentration was apparently not harmful. At higher concentration sodium chloride has enough chance to induce genetic disturbances. However, it needs further investigations in order to have a better understanding of the mechanism of the effect of sodium chloride on seedling growth of *V. radiata* L.

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