



Effect of Salt Stress on Seedling growth and Survival of *Oenothera biennis* L.

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Available online at: www.isca.in, www.isca.me

Received 26th June 2014, revised 6th August 2014, accepted 18th September 2014

Abstract

Effect of salinity on emergence, seedling growth and seed vigor of *Oenothera biennis* L., a medicinal plant was considered using different concentrations of NaCl (25 mM, 50 mM, 75 mM and 100 mM). Increasing stress regimes upto 50 mM led to enhancement of seed germination. At higher salt concentrations a negative relationship between seed germination was obtained. Seedling survived and grew up to salinity of 100 mM NaCl and eventually this species is tolerant to seedling stage. Elongation of stem and root was decelerated by increasing salt stress. Though, this species has a tendency for rapid root penetration and roots are able to draw water from saline soil. Seed vigor index decreased with increasing concentration of stress.

Keywords: Stress, germination, seedling growth, seed vigor index, evening primrose.

Introduction

At present salinity is the world-wide problem and is increasing day by day due to excessive use of chemical fertilizers and saline water for irrigation, especially in arid and semi arid areas. Salinity adversely affects the germination and survival of most of glycophytes. It is also well documented in literature¹⁻³. So understanding the morphological responses of plants to salinity are utmost importance. Germination of seeds is the first critical and most sensitive stage in life cycle of plants⁴ and the seeds exposed to unfavourable environmental conditions like salts stress may have to compromise the seedling establishment.⁵ Salinity either completely restrains germination at higher levels or induces state of dormancy at low levels⁶. However, plant species varies in their sensitivity or tolerance to salts⁷. The decrease in growth under salinity is a result of many physiological responses which include alteration of water status, photosynthetic efficacy, carbon allocation and utilization⁸.

Medicinal herbs have been extensively studied because chemical medicines have proved to have side effects and human tend to use natural products as much as possible⁹. Moreover, medicinal plants play a monumental role in the provision of health care in many developing countries¹⁰. The need is to study the medicinal plants for their halotolerance. *Oenothera biennis* L., an important medicinal plant, known as evening primrose is cultivated in Indian gardens¹¹. It is an exotic as it originated in North America¹². The plant is recognized as oil seed crop contains approximately 7-10 percent G-linolenic acid (GLA), an essential fatty acid. GLA has been identified as a useful treatment for a variety of ailments including blood pressure, cardiovascular disease, skin disorder and diabetic neuropathy¹³. The purpose of present study is to analyze the *Oenothera biennis* L. in terms of germination and seedling growth to salinity stress.

Material and Methods

Seed Source: Seeds of *Oenothera biennis* L., collected from Dr. Y. S. Parmar University of Horticulture and Forestry, Nauni, Solan (Himachal Pradesh) were used in this study. The experiment was conducted from April to May, 2013 under laboratory conditions at Shoolini University of Biotechnology and Management Sciences, Solan. The detailed schedule of study was as:

Seedling germination: Seeds of *Oenothera biennis* L., selected on the basis of size and colour, were surface sterilized with 0.1 percent HgCl₂ solution for 5 minutes followed by thorough washing with distilled water. Then, the seeds were soaked in distilled water (control) or in solutions of stated concentrations of effector namely 25, 50, 75, 100 mM NaCl solution for 24 hours. Afterwards, the seeds were shifted to petriplates lined with three layers of filter papers moistened either only by distilled water (control) or by effect of different NaCl concentrations of same volume. The treatments were replicated six times and each petriplate contained thirty seeds. The seeds were then allowed to germinate in an incubator at 25 ± 2°C under continuous illumination provided by fluorescent white light. Emergence of 2-5 mm radicle was taken as seed germination¹⁴. The seed germination was recorded at periodic intervals for few days until the final count.

Seedling growth and seed vigor: After 31 days of incubation, seedling growth was measured in terms of root length, shoot length, seedling fresh weight and seed-vigor index (SVI). The seed-vigor index (SVI) was determined with the help of Seghatoleslami method¹⁵: Germination percentage × means of seedling length (cm)/100. Each experiment was performed in triplicate; each replicate comprised thirty seeds. At the end of experiment, data was subjected to analysis of variance

(ANOVA). The statistical analyses were done using Graph Pad Prism® 5.2.

Results and Discussion

Effect of salinity on seedling emergence: Seedling started to emerge 3 days after sowing and 72 % seed germination was obtained over period of 31 days under control as presented in figure-1. Seedling emergence was noticed on the 3rd, 5th, 7th and 9th days after sowing in 25, 50, 75 and 100 mM NaCl, respectively and seed germination percentage was 76%, 73 %, 60.9% and 48% respectively. Seedling did not emerge from soils with further increase in salinity. There was significant reduction in germination of seed with increasing salt stress. As a result it has been observed that this plant species is salt tolerant at seed germination phase upto certain concentration. However, salt concentrations exceeding 50 mM NaCl were detrimental to seed germination. The diminishing germination due to raising salinity can be correlated to the nature of salinity to restrict imbibitions of water due to lowered osmotic potentials of the medium and causes changes in metabolic activity¹⁶. Different studies showed that different salinity stress levels affect germination in variety of plants such as *Satureja hortensis*¹⁷ and *Elymus junceus*¹⁸. High level of soil salinity can significantly suppress seed germination in glycophytes and halophyte plants. This suppression is because of potential osmotic effects and ionic toxicity¹⁹.

Effect of salinity stress on seedling growth: Shoot and root growth inhibition is a common response to salinity. However, shoots are usually sensitive to cation interference than roots and there are huge diversity among plant species in the capacity to avoid or tolerate the excess salt concentration²⁰. Shoot length of seedling given in figure-2(a) lessened with the increase of salt treatments. Salt stress inhibits the efficacy of the translocation

and assimilation of photosynthetic products²¹ and might have caused decrease in shoot growth. In the present study, increasing NaCl given in figure-2(b) resulted in decrease in root length at higher concentrations but at lower concentration of 25 mM NaCl root length increased in comparison to control. NaCl had a stimulatory effect on the growth of root upto a certain concentration. High concentration of salt results in slow down or stop root elongation²² and causes reduction in root production²³. Similarly in *Catharanthus roseus*; *Withania somnifera* and *Salvadora persica*, root length was reduced in comparison to untreated plants with increase in salinity²⁴, which is in consensus with the present study. However, in *Plantago ovata*, initially root first enhanced then significantly decreased with increasing concentrations of NaCl²⁵.

Seedling fresh weight in *O. biennis* L. presented in figure-2(c) also decreased in concentration dependent manner. It seems that the decreased seedling fresh weight may be because of decreased water absorption due to osmotic stress induced by NaCl, which in turn causes a reduction in the amount of water in plant tissue. Massai *et al.*,²⁶ observed that salinity decreased plant growth due of reduction in photosynthesis, closing of stomata and reduction of water entrance into the plant. In *Artemisia annua* L. seedling fresh weight decreased with increase in salinity. In *O. biennis* L. seed vigor index declines as shown in figure-3 with increasing NaCl concentration. The most seed vigor index was related to control treatment. Generally, seed vigor index is related to special influence of ions and reduction of environmental water potential in the presence of salinity. If salinity raises there is reduction of environmental osmotic potential and seed vigor index show negative trends²⁸. Liu and co-workers²⁹ opined that this might be due to suppression of cell division induced by chromosomal aberrations.

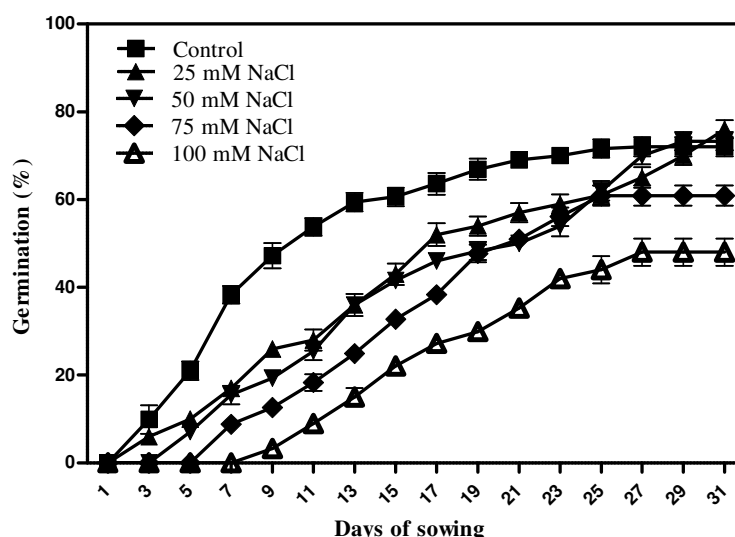


Figure-1
Time-course of germination of *Oenothera biennis* L. seeds as affected by NaCl. Values are mean \pm S.E., n=6

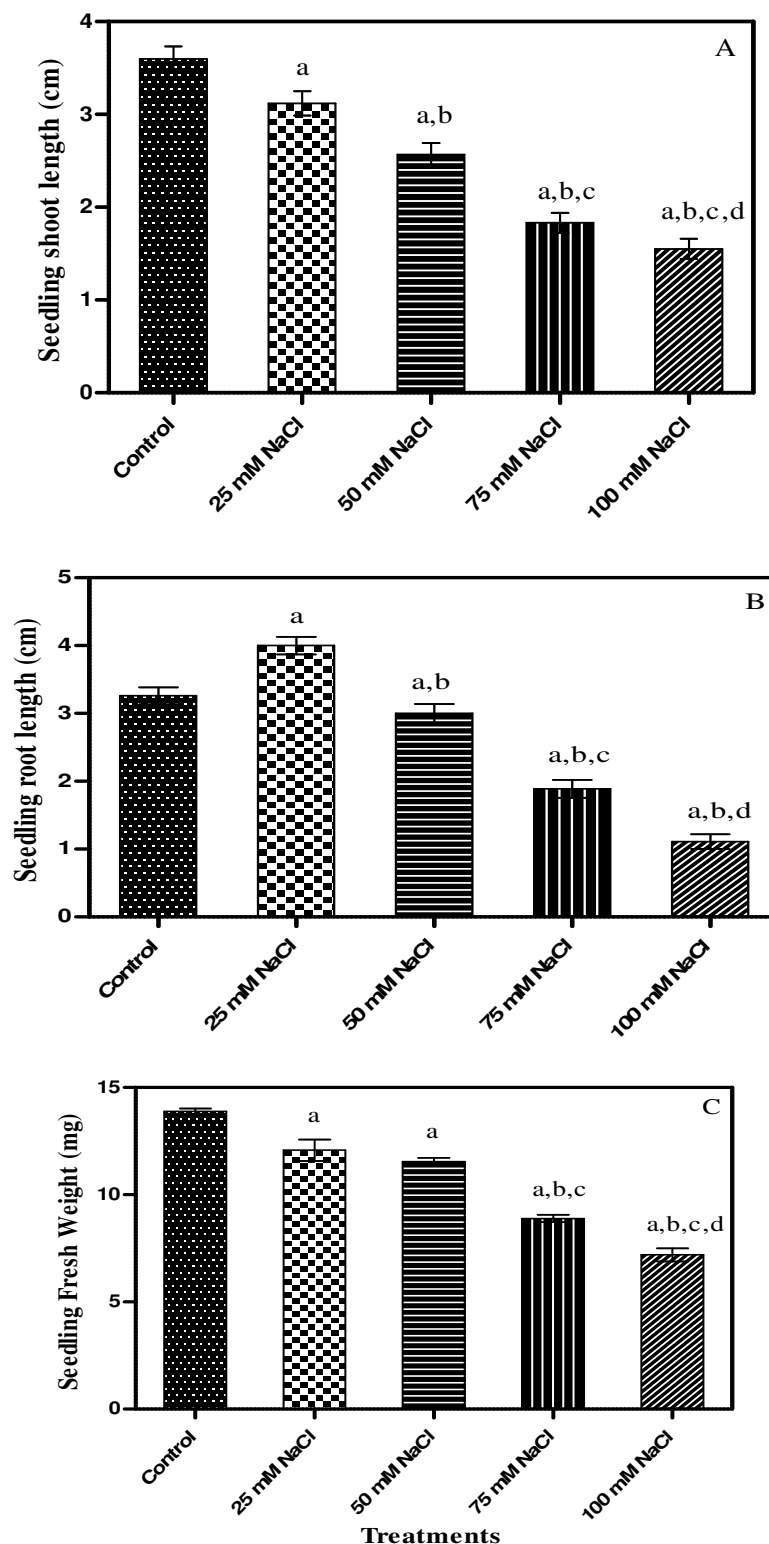


Figure-2

Effect of NaCl on seedling shoot length (A), root length (B) and seedling fresh weight (C) of *O. biennis* L. Values are mean \pm SE; n=6. Analyzed by One-way ANOVA followed by Tukey's multiple comparison test. ^ap<0.05 Vs control, ^bp<0.05 Vs 25 mM NaCl, ^cp<0.05 Vs 50 mM NaCl, ^dp<0.05 Vs 75 mM NaCl, ^ep<0.05 Vs 100 mM.

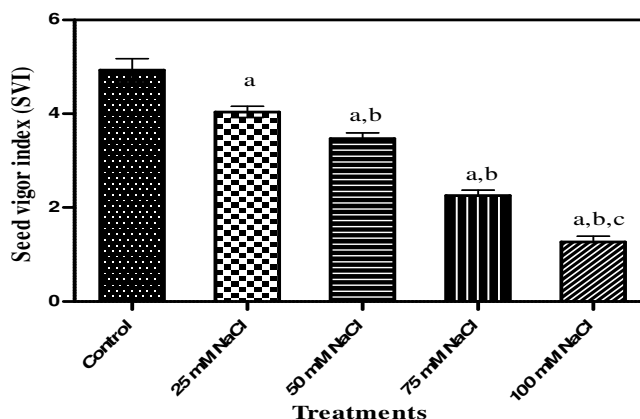


Figure-3

Seed vigor index (SVI) of *Oenothera biennis* L. under NaCl stress conditions. Values are mean \pm SE; n=6. ^ap<0.05 Vs control, ^bp<0.05 Vs 25 mM NaCl, ^cp<0.05 Vs 50 mM NaCl, ^dp<0.05 Vs 75 mM NaCl, ^ep<0.05 Vs 100 mM NaCl, analysed by One-way ANOVA followed by Tukey's multiple comparison test

Conclusion

Plants have inbuilt ability to adjust to seasonal environmental variables. Apart from the environmental variables, there may be certain other rapid and predictable environmental disturbances resulting in stressful conditions³⁰. However, plants differ in ability to tolerate such stressful situations. This study shows that salinity stress restricts the seedling growth and seed germination of *O. biennis* L., decreasing shoot length, root length, fresh weight at higher stress regimes. However, plant can grow well under mild stress conditions. The findings presented demonstrate that, in terms of the parameters being investigated and in comparison of previous reports available in the literature, *O. biennis* L. would be classified as a species tolerant to salinity during seedling growth at lower level of salinity.

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