# Effect of saline stress on growth and biochemical indices of chrysanthemum (Chrysanthemum morifolium) germplasm

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#### ABSTRACT

Chrysanthemum (*Chrysanthemum morifolium* Ramat.) is one of the most popular cut flowers in the world and occupies important position in the world flower industry. An experiment was conducted to study the effect of saline stress on growth and biochemical indices on 10 chrysanthemum germplasm. It was determined by performance comparisons against control tests (without NaCl treatment) which acts as an indicator. Germplasm selected for the experiment were Garden Beauty, Shanti, Red Stone, Basanti, Yellow Reflex, Arka Ravikiran, Anmol, Mother Teresa, Sweta Singar and Jaya. Salt stress had significant effects on plant growth, assimilator pigment content and proline concentration. With application of 150 mM NaCl, there is reduction of shoot growth, root growth, leaf area, leaf number, shoot dry weight and chlorophyll content, whereas level of proline increased with increase salinity. Shoot elongation was less sensitive to salinity than root growth, with significant reductions in root growth at high salinity levels (150 mM). The genotype-Garden Beauty (5.03%) recorded minimum shoot length reduction, whereas minimum shoot dry weight reduction was observed in Anmol (23.97%) with increasing salinity. The genotype Basanti (2.35%) recorded minimum reduction in root growth. Minimum reduction in photosynthetic pigments was observed in Mother Teresa (51.55%) and high accumulation of proline content was recorded in Sweta Singar (162.50%). The genotypes Mother Teresa, Anmol, Basanti and Sweta Singar were found to be tolerant due to enhanced photosynthetic pigments, proline concentration and better shoot and root growth with higher salinity.

**Key words:** Chrysanthemum, Germplasm, Salt tolerance, Screening

Salinity is known to affect one-third of the world's arable land. It is the major abiotic stress reducing the yield of wide variety of crops all over the world (Tester and Davenport 2003). Salt affect osmotic stress and ionic toxicity of plants, affecting all major plant processes (Yadav et al. 2011). Growth and yield performance reduced through a decrease in ability of plants to take up water from the soil solution and the destruction of soil structure (Barrett-Lennard 2003). Salinity tolerance of flowers crops is of increasing importance due to decreasing availability of high-quality irrigation water. Increasing the salt tolerance of crops-will allow the effective use of poor quality irrigation water. Currently, only few major determinant genetic traits of salt tolerance have been identified owing to low success in releasing of salt-tolerant crops for optimal use of saltcontaminated water resources (Flowers 2004, Munns 2005). Chrysanthemum (*Chrysanthemum morifolium* Ramat.) is the leading ornamental species of economic importance, and

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the environmental conditions especially salinity and drought highly define its yield and quality. Several approaches have been developed for abiotic stress tolerance but important ornamental crops like chrysanthemum were rarely tapped. Therefore, an attempt has been made to study the effect of saline stress on growth and biochemical indices of chrysanthemum germplasm.

#### MATERIALS AND METHODS

The present investigation was conducted at National Phytotron Unit, IARI, New Delhi during 2015-16. The effect of saline stress on the chrysanthemum germplasm was determined by performance comparisons against control tests (without Nacl treatment) which acts as an indicator. Ten chrysanthemum germplasms were selected for the experiment, viz. Garden Beauty, Shanti, Red Stone, Basanti, Yellow Reflex, Ravi Kiran, Anmol, Mother Teresa, Sweta Singar and Jaya. Suckers of chrysanthemum germplasm were kept in plug trays for rooting in shade net and transferred after sufficient root establishment. Nutrient solution was used in hydroponics system and rooted cuttings were kept in Hoagland solution for a week initially for acclimatisation. Treatments were given after a week using 0 mM NaCl (control) and 150 mM NaCl. Interaction effect of salinity and

germplasm was significant for the parameters studied. For each parameter measured, analysis were performed on raw data for three mean replicate tank values per treatment. All data were analyzed with SAS release 8.2 using proc GLM (SAS Institute 2001). A completely randomized design was used, each treatment was replicated three times. Observations for plant growth, viz. shoot length (cm), root length (cm), leaf area (cm<sup>2</sup>), leaf numbers and shoot dry weight was recorded at the end of the experiment. Chlorophyll and chlorophyll b content assays were performed according to Lichtenthaler (1987). The proline concentration was determined using fresh material according to Bates *et al.* (1973).

#### RESULTS AND DISCUSSION

Effect of salt on plant growth

Increment in salinity level cause decline in shoot growth, root development, leaf area and leaf number among the chrysanthemum germplasm (Table 1) (Fig 1). Shoot growth was significantly decreased by increase salinity level compared to control. Maximum reduction in shoot growth was observed in Sweeta Singar (26.67%) followed by Anmol (25.49%) when plants were treated with 150 mM NaCl compared to their performance in non-saline condition, whereas minimum decrease for the parameter was observed in Garden Beauty (5.03%) followed by Mother Teresa (6.92%). Hasanuzzaman et al. (2013) reported that plants are susceptible to salinity during the seedling and early vegetative growth stage. Natural hormones might be affected due to the saline conditions leading to unbalanced growth of the plant (Lewis et al. 1980). Effects of salinity in reducing growth of different ornamental plants have also been reported in gladiolus (Ahir and Singh 2017), in marigold (ValdezAguilar et al. 2009), in zinnia (Zivder et al. 2011) and in rosemary (Kiarostami et al. 2010). Salinty has significant effects on root growth of Chrysanthemum germplasms. Maximum reduction in root length was observed in Mother Teresa (42.86%) followed by Red Stone (26.88%) when plants were treated with 150 mM NaCl compared to their performance in non-saline condition, whereas minimum decrease for the parameter was observed in Basanti (2.35%) followed by Yellow Reflex (9.09%). High external salt concentrations can also inhibit root growth, with a reduction in length and mass of roots. Cassaniti et al. (2009) also reported on decreasing plant height and root growth with increase salinity in ornamental shrubs. The overall negative effect of salt on growth attributes in the

present study also conforms to the findings of Ashraf and Ahmad (2000) in cotton. Significant reduction in leaf area was observed at high salinity level in all the chrysanthemum germplasms. The genotype Shanti (24.11%) recorded maximum reduction in leaf area followed by Red Stone (17.09%) when plants were treated with 150 mM NaCl compared to their performance in non saline condition whereas minimum decrease in leaf area was observed in Garden Beauty (8.11%) followed by Mother Teresa (7.32%). At high NaCl concentration (150 mM), the germplasm shows chlorotic leaves whereas the leaves of controlled plants were comparatively green and fully expanded. Reduction in leaf number was also observed at high salinity level in all the germplasm and maximum reduction was observed in Jaya (70.00%) followed by Shanti (60.00%) when plants were

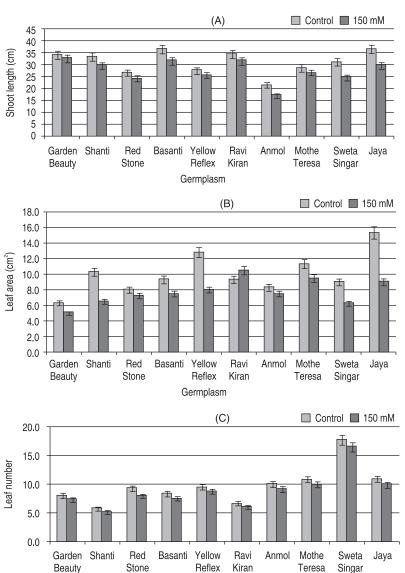


Fig 1 Effect of salt stress (0 and 150 mM NaCl) on plant growth of 10 Chrysanthemum germplasm: Shoot length (A), Leaf area (B), and Leaf number (C). Each value represents the mean of three replicates  $\pm$  SE. Twoway analysis of variance for the interaction of germplasm x treatment and main effects of germplasm and treatment were significant at P < 0.05.

Germplasm

Table 1 Root length and proline concentrations (mean±SE) in chrysanthemum germplasms subjected to salinity stress at different concentrations of NaCl

Germplasm	Root length (cm)		Proline (µmol/g FW)	
	NaCl (150 mM)	Control	NaCl (150 mM)	Control
Garden Beauty	$12.0 \pm 1.16$	$14.3 \pm 0.79$	$4.5 \pm 0.90$	$1.6 \pm 0.01$
Shanti	$11.5\pm0.54$	$13.0\pm1.25$	$18.3\pm3.32$	$1.2\pm0.02$
Red sone	$9.3 \pm 0.99$	$11.8\pm0.96$	$11.0 \pm 2.85$	$1.3 \pm 0.29$
Basanti	$8.5\pm0.68$	$8.7\pm0.47$	$13.4\pm2.16$	$0.9\pm0.01$
Yellow Reflex	$11.0 \pm 1.12$	$12.0 \pm 1.16$	$8.3 \pm 1.16$	$2.1 \pm 0.05$
Arka Ravikiran	$10.0 \pm 1.03$	$11.2 \pm 0.59$	$11.5 \pm 0.95$	$2.2 \pm 0.01$
Anmol	$6.0\pm0.66$	$7.3\pm0.64$	$18.2\pm2.98$	$1.2\pm0.69$
Mother Teresa	6.3±1.02	$9.0 \pm 1.31$	$13.1 \pm 0.94$	$1.5 \pm 0.04$
Sweta Singar	$8.3 \pm 0.14$	$9.8 \pm 0.75$	$14.0 \pm 0.90$	$2.6 \pm 0.46$
Jaya	$8.0\pm0.50$	$10.3\pm0.47$	$9.3 \pm 1.00$	$0.4\pm0.35$

treated with 150 mM NaCl compared to their performance in non-saline condition, whereas minimum decrease for the parameter was observed in Arka Ravikiran (3.20%) followed by Red Stone (10.34%). When plants are exposed to salinity at an early stage of seedling development, there are linear reductions in both leaf area expansion and total plant biomass. Leaf area reduction could be caused by a decrease in turgor in the leaves, as a consequence of changes in cell wall properties or a reduction in photosynthetic rate. Cassaniti *et al.* (2009) also reported that salinity reduced cell elongation and division in plant cell, thereby decreasing number of leaves and leaf area in ornamental shrubs.

## Effect of salt on shoot dry weight

Increment in salinity level significantly reduced shoot dry weight in the moderately sensitive chrysanthemum germplasm (Fig 2). Maximum reduction in shoot dry weight was observed in Garden Beauty (97.80%) followed by Basanti (93.55%) when plants were treated with 150 mM NaCl compared to their performance in non saline condition whereas minimum decrease was observed in Anmol (23.97%) followed by Mother Teresa (35.14%). High reduction for the parameters was also observed in Red Stone (92.42%) and Shanti (92.31%) and was statistically at par. Salt tolerance is marked by percent biomass production as growth takes place in saline condition when compared with non saline condition. Metabolic processes like enzyme activities, protein and nucleic acid synthesis might induced inhibitory effects on dry and fresh weight of shoots and roots with increased salinity. The results regarding plant biomass reported in the current study is in accordance with the findings Ltaief et al. (2007). Cassaniti et al. (2009) added in his findings that the decrease in shoot dry weight and leaf area were the first visible effects of salinity both in sensitive and tolerant species such as *Cotoneaster lacteus* and *Eugenia myrtifolia* respectively.

## Effect of salt on photosynthetic pigments

Increment in salinity level significantly reduced photosynthetic pigments in the moderately sensitive chrysanthemum germplasm (Fig 2). Maximum reduction in chlorophyll a content was observed in Arka Ravikiran (96.77%) followed by Anmol (93.65%) when plants were treated with 150 mM NaCl compared to their performance in non-saline condition, whereas minimum decrease was observed in Garden Beauty (10.75%) followed by Sweta Singar (16.39%). Maximum reduction of chlorophyll b content was observed in Shanti (99.16%) followed by Garden Beauty (80.00%) when plants were treated with 150 mM NaCl compared to their performance in non-saline condition, whereas minimum decrease was observed in Mother Teresa (51.55%) followed by Red Stone (52.36%). A decrease in the chlorophyll content with high salt concentration might be associated with disruption in cellular functions and damage to photosynthetic electron transport chain or membrane deterioration (Djanaguiraman et al. 2016). Zhong (1997) also reported that leaf chlorophyll contents of Nanjing Chrysanthemum indicum and Chrysanthemum chanetii increased with low salt concentration, but decrease with increment in salt concentration. Reducing effect of salinity on photosynthesis and water use efficiency was also reported by Nahed et al. (2011) on Matthiola incana and Azza et al. (2011) on Schefflera arboricola. The increased enzyme activity in salt stress is associated with decreased photosynthetic pigments (chlorophyll a, chlorophyll b and carotenoids) content as reported in Picea sp (Croser et al. 2001), and Rosmarinus officinalis (Kiarostami et al. 2010). In sweet cherry rootstock Gisela 5 (Prunus cerasus × Prunus canescens), a decrease in chlorophyll content was alao observed as a result of sodium chloride exposure (0, 50, 100, and 150 mM) (Erturk et al. 2007).

### Effect of salt on proline content

Increase salinity level significantly reduced the level of proline in the moderately sensitive chrysanthemum germplasm (Table 1). Maximum increase in proline concentration was observed in Sweta Singar (162.50%) followed by Basanti (98.57%) when plants were treated with 150 mM NaCl compared to their performance in non-saline condition, whereas minimum decrease of proline level was observed in Garden Beauty (16.96%) followed by Red Stone (23.35%). Proline accumulation in tissues of plants exposed to saline stress can be attributed to an increase in proline synthesis and a reduction in its degradation. Their deposition induced by salt stress leads to clearing of free radicals in cells which act as a tolerance mechanism of osmotic regulation (Bellinger et al. 1991). Oprica Lacramioara et al. (2015) reported that accumulation of proline in the marigold seedling treated with 100 mM NaCl was by 2.8 times higher than control. Similar result of enhanced proline

1.5

1.0

0.5

0.0

Garden

Beauty

Shanti

Red

Sone

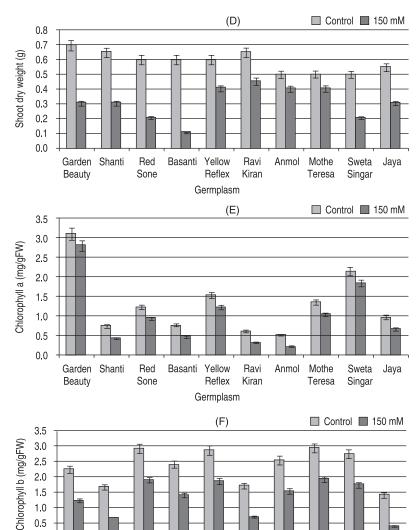


Fig 2 Effect of salt stress (0 and 150 mM NaCl) on shoot biomass and chlorophyll content 10 chrysanthemum germplasms: Shoot dry weight (D), Chlorophyll a (E), and Chlorophyll b (F). Each value represents the mean of three replicates  $\pm$  SE. Two-way analysis of variance for the interaction of germplasm  $\times$ treatment and main effects of germplasm and treatment were significant at

Yellow

Reflex

Germplasm

Ravi

Kiran

Basanti

Mothe

Teresa

Sweta

Singar

Java

Anmol

concentration with increasing salinity was also reported by Azza et al. (2011) on Schefflera arboricola plant and Nahed et al. (2011) on Matthiola incana plants. Lutts et al. (1996) also reported on the significant relationship between proline accumulation and plant salt tolerance in rice.

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