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Evaluation of *Chrysanthemum morifolium* varieties for salinity tolerance under hydroponic system

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ABSTRACT

Salinity tolerance of 22 varieties of chrysanthemum (*Chrysanthemum morifolium* Ramat.) was studied in National Phytotron Facility, ICAR-Indian Agricultural Research Institute, New Delhi at five different salinity levels (0, 50, 100, 150, 200 mmol/L). Salt tolerance was identified by measuring changes of different physiological as well as morphological parameters such as chlorophyll, proline, MSI (Membrane Stability Index), shoot length, root length, dry weight, etc. In the present study, it was found that the amount of chlorophyll a, chlorophyll b and total chlorophyll decreased with increase in the amount of salt concentration. At 100 mmol/L NaCl, the highest total chlorophyll content was found in Pusa Aditya, Haldighati, Lalit, Little Pink and Jaya, whereas lowest was observed in Pusa Sona, Yellow Gold and Himanshu. The rate of reduction in dry weight is dependent on the level of stress and variety. The greatest reduction in dry weight was found under 200 mmol/L NaCl treatment. The MSI reduced up to 47.64% under 200 mmol/L NaCl treatment from 85.65% of control. Reduction in MSI with increase in salinity may be due to high electrolyte leakage from cells of susceptible varieties. An increase in Proline content was observed under salt treated plants as compared to the plants under control. There was significant decrease in the root length with increase in the salt concentration. On the basis of modified standard evaluation score (SES) of visual salt injury, the 22 varieties can be grouped into tolerant, moderately tolerant, susceptible and highly susceptible with most of the varieties falling under the category of moderately tolerant. Little Pink, Lalit, Haldighati and Pusa Aditya falling under the tolerant category while Himanshu and Pusa Sona are grouped under susceptible and highly susceptible category respectively.

Key words: Hoagland Solution, Membrane Stability Index, Proline, Salinity

Chrysanthemum (*Chrysanthemum morifolium* Ramat.) is native to China and is one of the most important and popular ornamental worldwide. There are more than 30 000 chrysanthemum cultivars worldwide and about 3 000 are grown in China (Li and Shao 1990, Chen 2001). Area under chrysanthemum cultivation in India is 18.36 thousand ha (NHB 2012-13). It is mainly grown in the states of Andhra Pradesh, Karnataka, Maharashtra and Tamil Nadu. Mostly commercial varieties are susceptible to infection by a number of pests, diseases, and viruses that seriously reduce their ornamental quality (Nagata and deA ´vila 2000, Chung *et al.* 2001). In addition, abiotic stress such as salt stress, causes serious crop losses, reducing yield by more than half (Boyer 1982, Bray *et al*. 2000). The problem of soil salinity is due to continuous use of nutrient solution (fertilization) in greenhouse as well as use of reclaimed water for irrigation due to the limited supply of fresh water, especially in arid and semiarid regions of the world.

Salinity is a major environmental factor affecting the performance of many crop plants (Munns 2002). Salinity inhibits the growth and reduces the yield of many plants. Reduction in growth of plants under salt stress leads to necrosis of the leaves which leads to the reduction in photosynthetically active area (PAR). Therefore, understanding the physiology of salt tolerance in plants is very important for mitigating the problem of salinity. The general characteristic of saline soils is the occurrence of high concentrations of soluble salts or high exchangeable Na (ESP>15% with pH>8.5) which interferes with the normal plant growth. Because of its increased osmotic potential, the water in saline soils is less readily available to plants leading to a physiological water stress. On the other hand the excessive uptake of Cl^- and Na^+ results in nutrient imbalances and ion toxicity. The restriction of the uptake of chlorine and sodium in the roots and the accumulation of these ions in stems and leaves are therefore, important mechanisms in the tolerance to salt stress which depends

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on the permeability and selection of the roots to these ions during uptake.

The effect of salinity on floricultural crops has been investigated to a much lesser extent than other crops. A better understanding of salinity effect on plant growth, morphology and physiology can help to screen the tolerant varieties and genotypes that can be grown in the areas where salinity is a major problem thereby enabling the growers to grow floricultural crops in problem areas and increasing their income.

Many investigations have been done to document the effect of soil salinity on different crops in India but till now no systematic work has been started for screening and development of varieties of chrysanthemum that can be cultivated under saline condition. Existence of differences in salt tolerance not only amongst different species, but also within species (Munns *et al.* 2002). Therefore, the present study was carried out for screening and evaluation of the chrysanthemum varieties which will be helpful to uncover the mechanisms involved in salt tolerance in chrysanthemum, profiling of existing varieties with respect to salt tolerance and to breed salt tolerant varieties.

MATERIALS AND METHODS

The study was carried out during 2014-2015 at National Phytotron facility, ICAR-Indian Agricultural Research Institute, New Delhi. Screening and evaluation of 22 varieties was done under the glasshouse (at temperatures) 21°C and 18°C day and night temperatures respectively and relative humidity of 75%. The varieties that were screened are Maggi White, Karnal Pink, Pusa Chiraksha, Red Gold, Little Pink, Lalit, Dolly Orange, John Baber, Pusa Aditya, Yellow Gold, Gauri, Flirt, White Prolific, Shymal, Pusa Kesari, Jubilee, Neelima, Jaya, Lal Pari, Basanti, Himanshu, and Pusa Sona.

Rooted cuttings were raised from healthy mother plants and were kept in the glasshouse for one week before the initiation of salt treatment. Plastic trays filled with Hoagland nutrient solution (pH 6.8 and EC 2.5 dS/m) were used for the experiment. Salinity treatment of 0(control), 50, 100, 150 and 200 mmol/L were induced after a week. Plants were stressed under hydroponic culture for 48 days, salt treatment were renewed every 3 days. All the biochemical parameters were estimated using fully expanded 4th leaf counting from the apex. The different biochemical and physiological parameters that are studied are chlorophyll (a, b and total), dry weight, membrane stability index, proline, root length, shoot length. After 48 days of salt treatment, the plants were uprooted and dried in oven 60°C and the plant dry weight was measured. The experimental design was completely randomised design and statistical significance was tested by Duncan's New Multiple Range Test ($P<0.05$).

For visual screening modified standard evaluation score (SES) of salt injury (Method adapted from Gregorio *et al*. 1997) with certain modifications was used and indicated in the (Table 1).

Table 1 Modified standard evaluation score (SES) for visual salt injury

Score	Observation	Tolerance
1	Normal growth, no leaf symptoms	Highly tolerant
3	Nearly normal growth, but leaf tips or few leaves whitish and rolled	Tolerant
5	Growth severely retarded; most leaves rolled; only a few are elongating	Moderately tolerant
7	Complete cessation of growth; most leaves dry; some plants dying	Susceptible
9	Almost all plants dead or dying	Highly susceptible

RESULTS AND DISCUSSION

Chlorophyll a

Among various abiotic stresses, salt stress is one of the most important factors that limit the productivity and performance of crop plants. Salt stress in the plants leads to induction of many physiological and biochemical responses in plant cell system that are many times governed by polygenes. This induction of response is primarily due to osmotic stress, specific ion effect, nutrient deficiency, etc. In this process, plants have developed many mechanisms that will enable them to survive under various kinds of stress including the salt stress. Numerous metabolism

Table 2 Modified standard evaluation score (SES) for visual salt injury of chrysanthemum genotypes

Genotype	Salinity score	Reaction to salinity
Maggi White	5	Moderately tolerant
Karnal Pink	5	Moderately tolerant
Pusa Chitraksha	5.5	Moderately tolerant
Red Gold	5	Moderately tolerant
Little Pink	2.5	Tolerant
Lalit	3	Tolerant
Dolly Orange	5	Moderately tolerant
John Baber	5	Moderately tolerant
Haldighati	2.75	Tolerant
Pusa Aditya	2.5	Tolerant
Yellow Gold	5	Moderately tolerant
Gauri	5	Moderately tolerant
Flirt	5	Moderately tolerant
White Prolific	4.5	Moderately tolerant
Shymal	4.5	Moderately tolerant
Jublee	4.5	Moderately tolerant
Neelima	5	Moderately tolerant
Jaya	5	Moderately tolerant
Lal Pari	5	Moderately tolerant
Basanti	5.5	Moderately tolerant
Himanshu	8.5	Susceptible
Pusa Sona	9	Highly susceptible

including osmo-regulation, ion transport and protection from oxidative damage are critical in governing high salt tolerance (Chandan *et al*. 2006). In the present study it was found that the amount of chlorophyll a, chlorophyll b and total chlorophyll decreased with increase in the amount of salt concentration. Therefore, leaf chlorophyll content can be used as an important parameter for selecting salt tolerant varieties. Similar finding was reported by Guan *et al.* (2012) in comparing the stress effect of NaCl on two chrysanthemum species.

Among the treated plants the chlorophyll content was found to be maximum under 100 mmol/L NaCl and then declined under higher concentration. Under 100 mmol/L concentration, the highest chlorophyll a content was recorded in Pusa Aditya (0.97 mg/g), Lalit (0.96 mg/g), Haldighati (0.94 mg/g), Little Pink (0.92 mg/g), and Jaya (0.92 mg/g) under 100 mmol/L NaCl, whereas lowest chlorophyll was observed in Pusa Sona (0.59 mg/g) followed by Himanshu (0.61 mg/g) as shown in Table 3.

Table 3 Chlorophyll a content of chrysanthemum varieties under different salinity treatment

Variety	Chl a (mg/g)					
	Different concentrations of salt solution (mmol/L)					
	0	50	100	150	200	Mean
Maggi White	0.77^{defgh}	0.74^{cd}	0.79^{cd}	0.77ab	0.49^{cde}	0.713
Karnal Pink	0.75^{fgh}	0.75^{cd}	0.81^{bc}	0.79a	0.42^{ef}	0.705
Pusa Chitraksha	0.82^{abcde}	0.79^{bcd}	0.84^{bc}	0.80a	0.58^{b}	0.766
Red Gold	0.79 ^{cdefg}	0.80^{bc}	0.82^{bc}	0.78ab	0.60^{b}	0.759
Little Pink	0.73^{gh}	0.76^{cd}	0.92^{a}	0.68ef	0.50^{cdef}	0.682
Lalit	0.78^{cdefgh}	0.75^{cd}	0.96^{a}	0.65^{ed}	0.59^{cdef}	0.7
Dolly Orange	0.81^{bcdef}	0.77$_{bcd}$	0.75^{d}	0.72cb	0.51^{c}	0.712
John Baber	0.78^{cdef}	0.72^{d}	0.80^{cd}	0.78ab	0.43^{def}	0.703
Haldighati	0.72^{h}	0.79^{dcd}	0.94^{a}	0.58^{ed}	0.40^{fg}	0.687
Pusa Aditya	0w.79^{cdefg}	0.72^{d}	0.97^{a}	0.62^{d}	0.42^{ef}	0.704
Yellow Gold	0.84^{abc}	0.78^{bcd}	0.75^{d}	0.80^{a}	0.68^{a}	0.77
Gauri	0.76^{efgh}	0.72^{d}	0.81^{bc}	0.78^{ab}	0.49^{cde}	0.712
Flirt	0.78^{cdefgh}	0.74^{cd}	0.83^{bc}	0.77^{ab}	0.50^{cd}	0.725
White Prolific	0.81^{bcdef}	0.76^{cd}	0.82^{bc}	0.79^{a}	0.44^{cdef}	0.724
Shymal	0.74^{gh}	0.72^{d}	0.84^{bc}	0.76^{ab}	0.50^{cd}	0.712
Jublee	0.77^{defgh}	0.74^{cd}	0.79^{cd}	0.69^{c}	0.48^{cde}	0.694
Neelima	0.82^{abcd}	0.73^{cd}	0.86^{b}	0.79^{a}	0.46^{cdef}	0.733
Jaya	0.86^{ab}	0.84^{b}	0.92^{a}	0.72^{bc}	0.63^{ab}	0.794
Lal Pari	0.84^{abc}	0.78^{bcd}	0.86^{b}	0.74^{abc}	0.60^{b}	0.764
Basanti	0.82^{abcde}	0.75^{cd}	0.80^{cd}	0.79^{a}	0.47^{cdef}	0.726
Himanshu	0.88^{a}	0.93^{a}	0.61^{e}	0.53^{ef}	0.34^{g}	0.658
Pusa Sona	0.83^{abcd}	0.98^{a}	0.59^{e}	0.51^{f}	0.38^{fg}	0.662
Mean	0.796	0.776	0.822	0.71	0.488	

Chlorophyll b

The chlorophyll b content increased up to 150 mmol/L NaCl treatment but after that it decreased sharply at 200mmol/L. At 100 mmol/L salt concentration, Himanshu showed the highest chlorophyll b content (0.67 mg/g) followed by Pusa Sona (0.66 mg/g) while Haldighati and Pusa Aditya recorded 0.66 mg/g and 0.62 mg/g of chlorophyll b, respectively (Table 4). It is very interesting to note that the rate at which chlorophyll b decreases with increase in salt stress is very high in Himanshu and Pusa Sona (from 0.69 mg/g at 150 mmol/L to 0.40mg/g at 200 mmol/L NaCl treatment and from 0.70 mg/g at 150 mmol/L NaCl treatment to 0.4 6mg/g at 200 mmol/L respectively) whereas the rate of decrease in chlorophyll b content is very slow in Pusa Aditya and Haldighati (from 0.60 mg/g at 150 mmol/L to 0.59 mg/g at 200 mg/g and from 0.58 mg/g at 150 mmol/L to 0.57 mg/g at 200 mmol/L NaCl treatment (Table 4).

Table 4 Chlorophyll b content of chrysanthemum varieties under different salinity treatment

Variety	Chl a (mg/g)					
	Different concentrations of salt solution (mmol/L)					
	0	50	100	150	200	Mean
Maggi White	0.48^{e}	0.46^{b}	0.52^{b}	0.61^{bcd}	0.38^{cdefg}	0.49
Karnal Pink	0.49^{e}	0.48^{b}	0.50^{b}	0.59^{d}	0.33^{defg}	0.479
Pusa Chitraksha	0.50^{e}	0.49^{b}	0.51^{b}	0.61^{bcd}	0.34^{defg}	0.49
Red Gold	0.48^{e}	0.47^{b}	0.49^{b}	0.60^{cd}	0.33^{defg}	0.475
Little Pink	0.67^{ab}	0.49^{b}	0.53^{b}	0.44^{ef}	0.32^{efg}	0.491
Lalit	0.69^{a}	0.47^{b}	0.50^{b}	0.48^{e}	0.40^{cde}	0.509
Dolly Orange	0.51^{e}	0.50^{b}	0.52^{b}	0.62^{abcd}	0.36^{cdefg}	0.502
John Baber	0.53^{cde}	0.51^{b}	0.54^{b}	0.64^{abcd}	0.36^{cdefg}	0.517
Haldighati	0.68^{ab}	0.49^{b}	0.66^{a}	0.58^{fg}	0.57^{fg}	0.509
Pusa Aditya	0.69^{ab}	0.47^{b}	0.62^{a}	0.60^{g}	0.59^{g}	0.482
Yellow Gold	0.49^{e}	0.48^{b}	0.51^{b}	0.61^{bcd}	0.36^{cdefg}	0.491
Gauri	0.47^{e}	0.44^{b}	0.50^{b}	0.58^{d}	0.41^{cd}	0.48
Flirt	0.48^{e}	0.45^{b}	0.49^{b}	0.59^{d}	0.38^{cdefg}	0.479
White Prolofic	0.46^{e}	0.48^{b}	0.51^{b}	0.66^{abcd}	0.40^{cde}	0.502
Shymal	0.47^{e}	0.48^{b}	0.50^{b}	0.59^{d}	0.42^{bc}	0.494
Jublee	0.50^{e}	0.48^{b}	0.49^{b}	0.62^{abcd}	0.51^{a}	0.521
Neelima	0.52^{de}	0.49^{b}	0.52^{b}	0.61^{bcd}	0.39^{cdef}	0.507
Jaya	0.48^{e}	0.44^{b}	0.50^{b}	0.62^{abcd}	0.37^{cdefg}	0.482
Lal Pari	0.49^{e}	0.48^{b}	0.55^{b}	0.49^{abdc}	0.66^{ab}	0.534
Basanti	0.48^{e}	0.46^{b}	0.52^{b}	0.68^{abc}	0.40^{cde}	0.508
Himanshu	0.61^{abc}	0.62^{a}	0.67^{a}	0.69^{ab}	0.40^{cde}	0.598
Pusa Sona	0.60^{bcd}	0.64^{a}	0.66^{a}	0.70^{a}	0.46^{a}	0.632
Mean	0.535	0.491	0.537	0.588	0.388	

Total chlorophyll

The total chlorophyll content of all the varieties was found to be maximum under 150 mmol/L NaCl and then declined under higher concentration (Table 5). The varieties differed significantly both under control and salt stress treatment. The data presented in table 5 showed highest total chlorophyll content in Pusa Aditya (1.62 mg/g), Haldighati (1.60 mg/g), Lalit (1.46 mg/g), Little Pink (1.45 mg/g), and Jaya (1.42 mg/g) under 100 mmol/l NaCl, whereas lowest in Pusa Sona (1.25 mg/g), Yellow Gold (1.26 mg/g) and Himanshu (1.28 mg/g).

Under control conditions, total chlorophyll was found to be highest in variety Little Pink, Lalit, Haldighati, Pusa Aditya, Himanshu and Pusa Sona, whereas Shymal had the lowest total chlorophyll. It was found here that even the variety such as Himanshu and Pusa Aditya showed high amount of total chlorophyll but the rate at the amount of total chlorophyll decrease with increase in the salt concentration is very fast. This drastic change in the chlorophyll was observed when the salt concentration was increased from 150 mmol/L NaCl to 200 mmol/L NaCl.

Dry weight

The data presented in Table 6 exhibited that the dry weight of all the genotypes was significantly reduced in stressed plants in comparison to that of the control. The rate of reduction is dependent on the level of stress and the variety. The greatest reduction in dry weight was found under 200 mol/L NaCl treatment and in variety Himanshu. Amongst the treated plant, highest dry matter content was found to be under 50 mmol/L concentration of variety Dolly Orange (7 g) and lowest under 200 mmol/L of variety Himanshu (1.6 g). Under controlled condition, the highest dry weight was of the variety Dolly Orange (7.34 g). This reduction in the dry weight of the plants with increase in the concentration of salt treatment was reported by Guan *et al*. (2012). Similar results were also reported by Lee *et al*. (2008) in *Chrysanthemum morifolium.*

Table 5 Total chlorophyll content of chrysanthemum varieties under different salinity treatment

Variety	Total Chl (mg/g)					
	Different concentrations of salt solution (mmol/L)					
	0	50	100	150	200	Mean
Maggi White	1.25^{de}	1.20^{b}	1.31^{de}	1.38^{abc}	0.87^{def}	1.203
Karnal Pink	1.24^{de}	1.23^{b}	1.31^{de}	1.38^{abc}	0.75^{ghi}	1.185
Pusa Chitraksha	1.32^{bcde}	1.28^{b}	1.35^{bcde}	1.41^{abc}	0.92^{cde}	1.256
Red Gold	1.27^{cde}	1.27^{b}	1.31^{de}	1.38^{abc}	0.93^{cde}	1.233
Little Pink	1.40^{abc}	1.25^{b}	1.45^{bc}	1.12^{e}	0.82^{fghi}	1.173
Lalit	1.47^{a}	1.22^{b}	1.46^{b}	1.13^{e}	0.99^{efgh}	1.209
Dolly Orange	1.32^{bcde}	1.27^{b}	1.27^{e}	1.34^{bc}	0.87^{def}	1.214
John Baber	1.31^{bcde}	1.23^{b}	1.34^{bcde}	1.42^{abc}	0.79^{fghi}	1.22
Haldighati	1.40^{abc}	1.28^{b}	1.60^{a}	0.98^{e}	0.71^{i}	1.195
Pusa Aditya	1.48^{a}	1.19^{b}	1.62^{a}	0.95^{e}	0.72^{i}	1.186
Yellow Gold	1.33^{bcde}	1.26^{b}	1.26^{e}	$1.4^{abc}1$	1.04^{ab}	1.261
Gauri	1.23^{de}	1.16^{b}	1.31^{de}	1.36^{abc}	0.90^{cde}	1.192
Flirt	1.26^{de}	1.19^{b}	1.32^{de}	1.36^{abc}	0.88^{def}	1.204
White Prolific	1.27^{cde}	1.24^{b}	1.33^{cde}	1.45^{ab}	0.84^{efgh}	1.226
Shymal	1.21^{e}	1.20^{b}	1.34^{bcde}	1.35^{abc}	0.92^{cde}	1.206
Jublee	1.27^{cde}	1.22^{b}	1.28^{e}	1.31^{cd}	0.99^{bc}	1.215
Neelima	1.34^{bcde}	1.22^{b}	1.38^{bcde}	1.40^{abc}	0.85^{efg}	1.24
Jaya	1.34^{bcde}	1.28^{b}	1.42^{bcd}	1.34^{bc}	1.00^{abc}	1.276
Lal Pari	1.33^{bcde}	1.26^{b}	1.41^{bcd}	1.40^{abc}	1.09^{a}	1.298
Basanti	1.30^{bcde}	1.21^{b}	1.32^{de}	1.47^{a}	0.87^{def}	1.234
Himanshu	1.49^{a}	1.55^{a}	1.28^{e}	$1.2^{2}d$	0.74^{hi}	1.256
Pusa Sona	1.43^{ab}	1.62^{a}	1.25^{e}	1.21^{d}	0.96^{bcd}	1.294
Mean	1.332	1.266	1.359	1.298	0.876	

Table 6 Dry weight of chrysanthemum varieties under different salinity treatment

Variety	Dry weight (g)					
	Different concentrations of salt solution (mmol/L)					
	0	50	100	150	200	Mean
Maggi White	4.09^{kl}	3.95^{fghijk}	3.88^{hi}	3.69^{gh}	2.89^{fgh}	3.701
Karnal Pink	4.14^{jk}	3.46^{jkl}	3.39^{jk}	3.18^{hij}	3.00^{fg}	3.436
Pusa Chitraksha	4.31^{ij}	3.81^{ghijkl}	3.70^{ij}	3.68^{gh}	3.03^{efg}	3.707
Red Gold	5.04^{g}	5.06^{cde}	4.45^{efg}	4.25^{ef}	2.20^{hij}	4.203
Little Pink	4.44^{i}	4.75^{de}	4.38^{fg}	3.59^{gh}	3.50^{cdef}	4.134
Lalit	6.36^{c}	5.86^{bc}	5.62^{bc}	4.60^{de}	3.80^{bcde}	5.249
Dolly Orange	7.34^{a}	7.01^{a}	6.98^{a}	6.81^{a}	3.00^{fg}	6.23
John Baber	4.82^{h}	$4.00^{fghijkl}$	4.08^{gh}	4.00^{fg}	3.60^{bcdef}	4.1
Haldighati	4.77^{h}	4.58^{efgh}	4.46^{efg}	4.85^{cd}	3.29^{defg}	4.391
Pusa Aditya	3.35^{o}	3.36^{kl}	3.29^{k}	3.00^{ijk}	2.83^{fgh}	3.168
Yellow Gold	6.59^{b}	6.00^{b}	5.95^{b}	5.42^{b}	4.20^{abc}	5.632
Gauri	5.39^{f}	5.00^{de}	5.02^{d}	5.48^{b}	3.94^{bcd}	4.968
Flirt	4.32^{ij}	4.28^{efghij}	4.82^{de}	4.62^{de}	3.42^{cdef}	4.292
White Prolofic	3.89^{nml}	3.70^{hijkl}	3.62^{ijk}	3.55^{ghi}	2.87^{fgh}	3.528
Shymal	5.67^{e}	5.50^{bcd}	5.41^{c}	5.21^{bc}	4.33^{ab}	5.227
Jublee	6.10^{d}	5.50^{bcd}	5.61^{bc}	5.48^{b}	4.83^{a}	5.505
Neelima	4.78^{h}	4.50^{efghi}	4.38^{fg}	4.29^{def}	3.88^{bcd}	4.367
Jaya	4.91^{gh}	4.62^{efg}	4.50^{ef}	2.48^{k}	2.00^{ij}	3.703
Lal Pari	3.72^{n}	3.00^{l}	2.89^{l}	2.68^{jk}	2.60^{ghi}	2.979
Basanti	4.82^{h}	4.56^{efgh}	4.45^{efg}	4.41^{def}	3.00^{fg}	4.249
Himanshu	3.82^{nm}	3.50^{jkl}	3.45^{jk}	3.40^{hi}	1.60^{j}	3.155
Pusa Sona	3.94^{klm}	3.66^{ijkl}	3.48^{jk}	3.45^{ghi}	2.00^{ij}	3.309
Mean	4.848	4.531	4.447	4.188	3.176	

Membrane Stability Index (MSI)

Membrane stability index was found to be high for all the varieties under control conditions, whereas it decreases

Table 7 Membrane stability index of chrysanthemum varieties under different salinity treatment

Variety	MSI (%)					
	Different concentrations of salt solution (mmol/L)					
	0	50	100	150	200	Mean
Maggi White	86.59^{abcd}	82.71^{abc}	78.37^{a}	50.48^{j}	48.70^{bc}	69.372
Karnal Pink	85.35^{abcde}	83.75^{ab}	75.31^{abcd}	54.25^{i}	46.58^{bcdef}	69.052
Pusa Chitraksha	84.22^{cde}	81.72^{bcd}	76.02^{abc}	58.77^{h}	44.65^{def}	69.079
Red Gold	82.33^{e}	80.59^{bcd}	77.57^{ab}	60.54^{gh}	45.66^{cdef}	69.341
Little Pink	86.18^{abcd}	85.42^{a}	69.66^{g}	65.71^{bcd}	58.48^{a}	73.093
Lalit	87.83^{ab}	85.85^{a}	71.78^{efg}	62.13^{efg}	59.34^{a}	73.387
Dolly Orange	86.33^{abc}	81.84^{bcd}	77.75^{a}	61.61^{efgh}	49.60^{b}	71.431
John Baber	82.35^{e}	80.78^{bcd}	74.20^{bcde}	62.28^{efg}	47.45^{bcde}	69.415
Haldighati	85.27^{abcde}	81.55^{bcd}	76.37^{abc}	66.16^{bc}	59.78^{a}	73.83
Pusa Aditya	87.66^{abcd}	80.72^{bcd}	78.47^{a}	70.17^{a}	60.30^{a}	75.465
Yellow Gold	84.36^{de}	81.38^{bcd}	76.57^{abc}	70.23^{a}	44.59^{def}	71.431
Gauri	85.56^{abcde}	78.58^{d}	72.45^{defg}	65.88^{bcd}	46.69^{bcdef}	69.835
Flirt	86.62^{abcd}	81.52^{bcd}	70.78^{efg}	64.38^{bcdef}	49.71^{a}	70.604
White Prolific	87.35^{abc}	82.61^{abc}	71.62^{efg}	61.44^{fgh}	44.31^{ef}	69.467
Shymal	83.55^{ed}	80.43^{bcd}	73.41^{cdef}	62.68^{defg}	45.19^{cdef}	69.055
Jublee	87.74^{abc}	79.41^{cd}	70.63^{gf}	66.62^{b}	46.22^{bcdef}	70.129
Neelima	88.45^{a}	81.51^{bcd}	70.66^{gf}	64.75^{bcdef}	46.21^{bcdef}	70.318
Jaya	85.57^{abcde}	82.75^{abc}	72.52^{defg}	64.15^{bcdef}	48.09^{bcd}	70.619
Lal Pari	84.66^{bcde}	80.61^{bcd}	71.65^{efg}	62.95^{cdefg}	45.27^{cdef}	69.033
Basanti	85.51^{abcde}	83.79^{ab}	70.17^{gf}	63.78^{bcdef}	43.13^{f}	69.28
Himanshu	84.44^{bcde}	81.65^{bcd}	76.31^{abc}	64.97^{bcde}	35.14^{g}	68.506
Pusa Sona	86.34^{abcd}	82.59^{abc}	70.17^{gf}	60.65^{gh}	33.05^{g}	66.563
Mean	85.651	81.902	73.751	62.938	47.646	

sharply with increase in the level of salinity. The MSI was reduced upto 47.64% under 200 mmol/L NaCl treatment from 85.65% of control (Table 7). Under 50 mmmol/L NaCl treatment highest MSI was found to be variety in Lalit (85.85%) and Little Pink (85.42%) and lowest in Gauri (78.58%). Under 200 mmol/L NaCl treatment highest MSI was observed in variety Pusa Aditya (60.30%) followed by Haldighati (59.78%), Lalit (59.34%) and Little Pink (58.48%), which may be due to less leakage of solutes from the cell.

The presence of high NaCl leads to change in the membrane permeability which is expressed by an increase in the solute leakage. The probable cause for this leakage of the solutes may be due to increased accumulation of H_2O_2 and Lipid peroxidation (Dionisio-Sese and Tobita 1998). Whereas lowest MSI was recorded in variety Pusa Sona (33.05%) followed by Himanshu (35.14%). Ghoulam *et al*. (2002) reported that electrolyte leakage was greater as salt concentration increased and reached the maximum values at 200 mmol/L NaCl when they compared the effects of salt stress on 5 beet cultivars. The leakage was higher in susceptible cultivars of sugar beet than tolerant ones. High electrolyte leakage means low Membrane Stability Index (MSI), similar results were obtained from present study.

Proline

An increase in Proline content was observed under salt treated plants as compared to the plants under control (Table 8). Similar results were reported by Zhu *et al*. (2013) while evaluating intergeneric hybrids under salt stress. On an average of all the treatments Pusa Aditya had accumulated maximum proline content (an average of 3.22 mg/l) followed by Lalit (3.20 mg/l), Little Pink (3.19 mg/l), Haldhighati (3.17mg/l). Guan *et al*. (2012) reported an increase in the leaf proline content under salt stress, while comparing the salt tolerance of three species. It was found that the accumulation of proline was high in *C. okiense* as compared to the *C. ornatum* and they concluded that *C. okiense* is more salt tolerant than C. *ornatum*. The accumulation of osmolytes such as proline will help in the maintenance of structure and function of cellular macro-molecules and also helps to maintain high relative water content which will enable the plants to survive under stress condition.While proline content was found to be minimum in variety Pusa Sona (2.34 mg/l) followed by Himanshu (2.48 mg/g).The data presented in Table 8 revealed that the overall accumulation of proline highest under 50 mmol/l NaCl treatment (3.99 mg/g) followed by 100 mmol/l 3.47 mg/g and lowest under control condition (1.38 mg/g).

Table 8 Proline content of chrysanthemum varieties under different salinity treatment

Variety	Proline (mg/L)					
	Different concentrations of salt solution (mmol/L)					
	0	50	100	150	200	Mean
Maggi White	1.58^{gh}	4.43^{bcdef}	3.00^{g}	2.82^{cd}	2.72^{c}	2.913
Karnal Pink	1.56^{h}	4.26^{cdefg}	2.96^{g}	2.63^{de}	2.64^{f}	2.813
Pusa Chitra ksha	2.21^{b}	4.80^{abc}	3.56^{e}	2.94^{c}	2.04^{m}	3.113
Red Gold	3.04^{a}	4.94^{ab}	3.02^{g}	2.59^{e}	2.21^{k}	3.163
Little Pink	1.98^{c}	2.83^{h}	4.58^{b}	3.80^{a}	2.75^{e}	3.1912
Lalit	1.71^{def}	3.06^{h}	4.92^{a}	3.88^{a}	2.40^{i}	3.2
Dolly Orange	2.04^{c}	5.06^{a}	4.23^{c}	3.86^{a}	2.94^{c}	3.629
John Baber	1.68^{efg}	4.01^{efg}	2.98^{g}	2.05^{f}	1.84^{o}	2.515
Haldighati	2.02^{c}	2.07^{i}	4.09^{c}	3.85^{a}	3.84^{a}	3.1752
Pusa Aditya	1.56^{h}	2.84^{h}	4.54^{b}	3.88^{a}	3.27^{b}	3.2204
Yellow Gold	1.64^{efgh}	4.21^{defg}	2.98^{g}	2.40^{e}	2.32^{j}	2.713
Gauri	1.60^{fgh}	3.72^{g}	3.32^{f}	2.46^{e}	2.21^{k}	2.666
Flirt	1.70^{def}	4.56^{abcd}	3.29^{f}	2.96^{c}	2.54^{g}	3.013
White Prolofic	2.04^{c}	3.89^{fg}	3.01^{g}	2.84^{cd}	2.49^{h}	2.856
Shymal	1.80^{e}	3.88^{fg}	3.01^{g}	2.96^{c}	2.45^{ih}	2.823
Jublee	1.74^{de}	4.05^{efg}	3.26^{f}	2.40^{e}	2.08^{m}	2.709
Neelima	1.94^{c}	3.95^{fg}	3.54^{e}	3.20^{c}	2.88^{d}	3.105
Jaya	1.98^{c}	4.65^{abc}	3.89^{d}	2.94^{c}	2.86^{d}	3.267
Lal Pari	1.54^{h}	4.44^{bcdef}	3.03^{g}	2.50^{e}	2.42^{i}	2.79
Basanti	1.68^{efg}	3.81g	2.96g	2.38e	2.15^{l}	2.599
Himanshu	1.36^{h}	3.71^{g}	3.36ef	2.06^{f}	1.94^{n}	2.488
Pusa Sona	1.38^{i}	3.07^{h}	2.84g	2.56^{e}	1.88^{o}	2.3484
Mean	1.577	3.993	3.475	2.955	2.497	

Table 9 Root length of chrysanthemum varieties under different salinity treatment

Variety	Root length (cm)					
	Different concentrations of salt solution (mmol/L)					
	0	50	100	150	200	Mean
Maggi White	8.0^{ab}	8.00^{a}	9.33^{a}	7.66^{abc}	6.66^{ab}	7.933
Karnal Pink	7.0^{ab}	8.00^{a}	8.00^{abc}	6.33^{abc}	5.66^{ab}	7
Pusa Chitraksha	6.0^{ab}	9.00^{a}	8.00^{abc}	8.00^{abc}	8.00^{ab}	7.8
Red Gold	5.0^{b}	8.00^{a}	8.00^{abc}	6.00^{bc}	6.00^{ab}	6.6
Little Pink	9.0^{a}	8.00^{a}	9.00^{a}	9.00^{a}	8.00^{ab}	8.6
Lalit	8.0^{ab}	8.00^{a}	9.00^{a}	8.00^{abc}	8.00^{ab}	8.2
Dolly Orange	7.0^{ab}	8.00^{a}	5.00^{c}	8.00^{abc}	5.33^{b}	6.667
John Baber	6.0^{ab}	8.00^{a}	7.00^{abc}	5.33^{c}	5.33^{b}	6.333
Haldighati	8.0^{ab}	8.00^{a}	8.00^{abc}	8.00^{abc}	8.33^{a}	8.067
Pusa Aditya	7.0^{ab}	8.00^{a}	9.00^{a}	8.00^{abc}	7.00^{ab}	7.8
Yellow Gold	8.0^{ab}	8.00^{a}	7.00^{abc}	8.00^{abc}	5.33^{b}	7.267
Gauri	8.0^{ab}	8.00^{a}	7.00^{abc}	8.00^{abc}	5.66^{ab}	7.333
Flirt	7.0^{ab}	9.00^{a}	8.00^{abc}	8.00^{abc}	7.00^{ab}	7.8
White Prolofic	6.00^{ab}	8.00a	8.00^{abc}	8.00^{abc}	7.00^{ab}	7.4
Shymal	5.66^{b}	7.00^{ab}	7.00^{abc}	8.00^{abc}	6.00^{ab}	6.733
Jublee	8.00^{ab}	6.00^{ab}	8.00^{abc}	8.00^{abc}	6.00^{ab}	7.2
Neelima	8.00^{ab}	8.00^{a}	9.00^{a}	8.00^{abc}	6.66^{ab}	7.933
Jaya	7.00^{ab}	9.00^{a}	7.00^{abc}	8.00^{abc}	6.00^{ab}	7.4
Lal Pari	8.00^{ab}	8.00^{a}	8.00^{abc}	6.00^{bc}	8.00^{ab}	7.6
Basanti	8.00^{ab}	8.00^{a}	8.66^{ab}	8.66^{ab}	8.00^{ab}	8.267
Himanshu	7.00^{ab}	4.00^{b}	8.00^{abc}	5.33^{c}	6.00^{ab}	6.067
Pusa Sona	7.00^{ab}	6.00^{ab}	5.33^{bc}	6.00^{bc}	5.33^{b}	5.933
Mean	7.212	7.727	7.788	7.47	6.606	

Root length

Roots are the most sensitive organ and are affected first under salt stress. There was significant decrease in the root length with increase in the salt concentration. The root length of 22 varieties was found to be decreased from 7.21 cm (mean of all varieties) under control to 6.60 cm under 200 mmol/L NaCl treatment (Table 9). Plants under control condition exhibited better root growth. Similar results were reported by Hossain *et al.* (2004). At 50 mmol/L NaCl treatment the maximum root length was found to be of variety Pusa Chitraksha, Flirt and Jaya (9 cm each) and minimum was found to be of variety Himanshu (4cm) followed by Pusa Sona and Jubilee (6 cm). At 200 mmol/l NaCl treatment the maximum root length was observed in variety Haldighati 8.3 cm followed by Lalit, Little Pink, Pusa Chitraksha, Lal Pari and Basanti (8 cm), whereas Pusa Sona has found to have minimum root length (5.33 cm) followed by Himanshu (6 cm) this reduction in the root growth may be due to the imbalance of certain nutrients and hormones at high salt concentration which are necessary for the development and growth of roots.

Shoot length

Data presented in Table 10 showed that the shoot length of all the varieties, was reduced significantly from 19 cm (maximum) under control to 8 cm (minimum) under 200 mmol/l NaCl treatment. But there is not much significant difference among the varieties for shoot length. Under control condition Red Gold was found to have maximum shoot length, whereas Flirt, Shymal and Lal Pari had minimum shoot length. At 50 mmol/l NaCl Pusa Chitraksha has maximum and Shymal has minimum shoot length. The shoot length of Maggi White (15.33 cm) was minimum and Haldighati (21.66cm) and Pusa Aditya (21.88 cm) were maximum at 100 mmol/l NaCl treatment. Similarly under 150 mmol/NaCl treatment Himanshu and Karnal Pink had shortest shoot length, whereas Lal Pari and Basanti had longest shoot length. At 200 mmol/l NaCl treatment White Prolific has maximum shoot length followed by Karnal Pink and Dolly Orange, whereas Himanshu (8cm) had minimum shoot length followed by Pusa Sona (10cm). This kind of decrease in the shoot length with increase in the salt concentration was reported by Lee *et al*. (2008).

Table 10 Shoot length of chrysanthemum varieties under different salinity treatment

Variety	Shoot length (cm)					
	Different concentrations of salt solution (mmol/L)					
	0	50	100	150	200	Mean
Maggi White	18.00^{a}	18.33^{abc}	15.33^{c}	14.33^{defg}	9.66^{gh}	15.133
Karnal Pink	16.00^{a}	20.33^{a}	18.33^{abc}	13.33^{fg}	18.33^{ab}	17.267
Pusa Chitraksha	15.00^{a}	21.33^{a}	16.33^{bc}	15.66^{bcdef}	11.33^{efgh}	15.933
Red Gold	19.00^{a}	18.33^{abc}	17.33^{abc}	18.33^{abcd}	16.33^{abcd}	17.867
Little Pink	18.33^{a}	17.33^{abc}	16.00^{bc}	15.33^{cdef}	12.33^{defgh}	15.867
Lalit	14.33^{a}	16.33^{abcd}	18.00^{abc}	19.00^{abc}	16.33^{abcd}	16.8
Dolly Orange	16.33^{a}	17.33^{abc}	19.33^{abc}	18.33^{abcd}	18.33^{ab}	17.933
John Baber	15.33^{a}	14.33^{bcd}	20.33^{ab}	19.66^{ab}	18.33^{ab}	17.6
Haldighati	16.33^{a}	17.33^{abc}	21.66^{a}	18.33^{abcd}	15.00^{bcde}	17.733
Pusa Aditya	18.33^{a}	17.33^{abc}	21.88^{a}	17.66^{abcde}	15.00^{bcde}	16.976
Yellow Gold	17.33^{a}	19.33^{ab}	19.33^{abc}	17.33^{abcdef}	18.33^{ab}	18.333
Gauri	16.33^{a}	14.33^{bcd}	17.66^{abc}	14.00^{efg}	13.00^{cdefg}	15.067
Flirt	14.33^{a}	16.33^{abcd}	15.33^{c}	16.33^{abcdef}	14.33^{bcdef}	15.333
White Prolofic	15.33^{a}	18.33^{abc}	19.33^{abc}	19.33^{abc}	20.33^{a}	18.533
Shymal	14.33^{a}	13.33^{cd}	18.33^{abc}	18.33^{abcd}	17.00^{abc}	16.267
Jublee	15.33 ^{a}	17.33^{abc}	17.33^{abc}	14.33^{defg}	10.00^{fgh}	14.867
Neelima	15.00^{a}	18.66^{ab}	20.33^{ab}	17.33^{abcdef}	16.33^{abcd}	17.533
Jaya	16.33^{a}	18.33^{abc}	21.33^{a}	19.00^{abc}	18.33^{ab}	18.667
Lal Pari	14.33^{a}	18.33^{abc}	18.33^{abc}	20.00^{a}	18.33^{ab}	17.867
Basanti	17.33^{a}	19.00^{ab}	19.66^{abc}	20.00^{a}	15.00^{bcde}	18.2
Himanshu	15.00^{a}	17.33^{abc}	19.66^{abc}	10.66^{g}	8.00^{h}	14.133
Pusa Sona	18.33^{a}	16.33^{abcd}	18.00^{abc}	14.00^{efg}	10.00^{fgh}	15.333
Mean	16.197	17.273	18.60045	16.84836	15	

Similar reports were also reported in many other crops such as tomato (Montesano and van Iersel 2007)

Excessive amounts of salt in plants can become toxic in older leaves, causing premature senescence and a reduction in total photosynthetic leaf area (Munns 2002). This reduction in leaf area results in low rate of plant photosynthesis and thus finally affects the shoot growth.

The present study revealed the existence of significant differences in salinity tolerance among the 22 genotypes. The tolerant genotypes adapt different mechanisms for salinity tolerance. According to modified standard evaluation score (SES) for visual salt injury, the chrysanthemum genotypes has been categorised into tolerant, moderately tolerant, susceptible and highly susceptible groups. Most of the varieties fall under the category of moderately tolerant. Little Pink, Lalit, Haldighati and Pusa Aditya are grouped under the tolerant category while Himanshu and Pusa Sona are grouped under susceptible and highly susceptible categories, respectively. The results indicate that screening at initial stage of plant growth under controlled condition is an effective technique which saves a lot of resources and man power.

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