



Effectiveness of various salinity on leaf growth of *Gazania*

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Abstract

Salt stress is induced by NaCl and it minimizes the growth owing to variation in interior plant structure. The present work was designed in line to investigate the behavior of *Gazania harlequin* (L.) seedlings were raised with dissimilar salt combinations (25 ppm, 50 ppm, 75 ppm & 100 ppm of NaCl solution). The CRD experiment with 4 repetitions was done. The NaCl treatments were repeated in 4 times with 20 days interval totally for 80 days after transplanting in pots. Anatomical observations were recorded by the microscope of in NaCl affected growth leaf. The results illustrated that salinity levels had negative effect on anatomical characters as (xylem region, phloem region, cortex region, epidermis region and density of leaf lamina). So, it is concluded from study that control treatment (without salinity effect) showed improved outcome comparatively others treatments which were under saline situation. As the salinity increases ultimately it affects leaf anatomy of plant.

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Introduction

Ecological aspects affect the characters, components and growth of living entities e.g. plants and its communities. If any of these factors go beyond the optimal tolerance in plant, consequently it forms salinity situation, which change its developmental structure, physiological and biochemical functions. Salinity is one type of environmental stresses (Lawlor, 2002). Salinity is severely affecting agricultural productivity and accretion of salts has damaging effect on agricultural soils that has been developed into vital ecological concerns (Jaleel *et al.*, 2007). Land is becoming non productive yearly due to its internal salt addition. The pivotal influence of salinity is retardation of crop production by reducing the release of hormone from lower parts to upper parts (Azooz *et al.*, 2004). Salt causes change an extensive range of metabolism, excessive stunted development, and diminished enzyme actions and biochemical compositions (Muthukumarasamy & Panneerselvam, 1997). Salinization acts as chief responsibility in soil deterioration. It disturbs 19.5 % of irrigated soil and 2.1 % of dry soil of agriculture. Along with different biotic stresses, salinity is the foremost cause that minimizes or damages the plant development, declines germination and at last lower down the crop production. Soil salinity is a state which indicated by immense composition of soluble salts.

Agriculture input in Pakistan's GDP is 20.6 % (Anonymous, 2009). At present, saline soils comprise of almost 11 m ha i.e. about 55 % of total arable soils leaving dangerous effects on national economy and retards national productivity (Sahi, 2006; FAOSTAT, 2008). Soil salinity and drought are hurdling plant production round the world (Pitman and Lauchli, 2002; Tanji, 2002; Hu and Schmidhalter, 2005). The overall worldwide range of soils damaged by salinization has been reported to be round about 830 million hectares (Martinez-Beltran and Manzur, 2005). In Pakistan, it is estimated that out of 6.67 million hectares saline patches exists on area of 222 million hectares in Punjab, 2.11 million hectares in Sindh, 2.31 million hectares in Baluchistan and 0.4 million hectares in KPK (Anonymous, 2005).

Ornamental plants can be considered all the species and diversity that present aesthetic pleasure, boost the atmospheric beautification and the nature of our lives (Save, 2009). Mostly the showy plants are glycophytes and diverge as susceptible to resistant against salt. The harmful outcome of salinity on development was presented by various researchers on diff. plants (Bry *et al.*, 2000, Wu *et al.*, 2007). Tremendous salinity levels have an effect on all facets of plant growth and plant accustomed to their surroundings by complex regulatory set-up (Gong *et al.*, 2001). Decline in growth and yield traits might take place owed to a decrease in capability of plants to uptake of water from the soil media and the devastation of soil composition (Barrett-Lennard, 2003). Salt resistant plants have the ability of good production because of uptake more water in such condition under saline soil while in most cases salinity retards the growth attributes resources required by salinity as there is an inadequate accessibility of pure water and low permeability in soil (Ashraf and Yasmin, 1997).

Gazania harlequin (L) is showy and decorative plant which is grown in parks. So, these annual ornamental are generally used for decoration purpose in diff. places (Vicki and Chan, 2008). Its blossoming starts in diff. times (early, mid) and in a few varieties in late summer. More sun light is compulsory for vigorous plants (Edward and Howe, 1999). Salinity creates internal variations in plant body and is capable of minimizing detrimental effects of salt stress (Poljakoff-Mayber, 1988). The pessimistic result of salinization on production was reported by (Hussein *et al.*, 2010; Abdel-Monem *et al.*, 2010) and (Saffan, 2008). If we want to face the challenges of 21st century we should have to devote interest to increase the yield of crops by developing salt tolerant cultivars (Ashraf, 2002). Most of researches have been conducted for salinity effect evaluation in main crops but there is tiny information in ornamental crops so, the attention is needed to check the salinity variation in ornamental plants. The study was taken in to judge the salinity influence on leaf anatomy in *Gazania*.

Materials and methods

An experiment in pots was held at Floriculture Research Area, UAF, Faisalabad during year (2012-13) in order to investigate the effects of salinity on development and flowering quality of *Gazania*. The CRD experiment with 4 repetitions was done.

After one month the seedlings of *Gazania harlequin* (L) was transplanted in the pots (20cm diameter and 22cm depth) containing a mixture of silt and leaf manure in 1:1. These seedlings were allowed to establish for 20 days before the start of applying diff. salinity levels (25 ppm, 50 ppm, 75 ppm, 100 ppm of NaCl solution). The soil wt. was measured and 1.5 kg (growth medium) put in all pots and the salinity zones were made artificially by adding up the solution of 25 ppm, 50 ppm, 75 ppm, and 100 ppm NaCl salt and one was as a control.

Statistical Analysis

For statistical analysis standard procedure was followed to record the data. Data collected was analyzed statistically using Fisher' (ANOVA) techniques (Steel *et al.*, 1997). The data was inserted for statistical analysis of variances procedure and the values of L.S.D were obtained (Snedecore & Cochran, 1980).

Anatomical Parameter

Leaf anatomy

Developed plants were detached from the soil and their whole leaf from plant was collected on the visual observation for anatomical study. Collected leaves were cleaned with distilled H₂O. After final washing, leaf was dipped into 70 % ethanol solution for fifteen days and their sectioning was done. After washing dipped it into 70 percent ethanol (C₂H₆O) solution for two weeks and their sectioning was done. Sections stained with safranine and fast green by follow the method of (Corgen & Widmayer, 1971). After sectioning, we prepared five different solutions of ethanol (C₂H₆O) for staining the slides to take the pictures of leaf. Sections were fixed in Canada balsam anatomical study under microscope and were photographed. The anatomical observations recorded during the study were the area of xylem, phloem, cortex, epidermis and lamina density of leaf in (µm²) according the methods described by (Johansen, 1940).

Results

Effectiveness of salinity on leaf growth of Gazania

Salinity showed adverse effect on leaf anatomy of *Gazania*. The statistical investigation showed the boost in salinity level there was significantly changes in different anatomical characteristics of leaf xylem region. Leaf area decreased time after time but considerably with increase in NaCl levels and was more affected than the Salt Range.

Table 1. Effect of different salinity levels on leaf anatomy of *Gazania* Same letter means don't vary significantly at P<0.05.

Treatment	PARAMETERS				
	leaf xylem area (µm ²)	leaf phloem area (µm ²)	leaf cortex area (µm ²)	leaf epidermis area (µm ²)	Leaf lamina thickness (µm ²)
Control	178537.81a	184870.61a	1634814.91a	329181.49a	5866.06a
25 ppm	123994.42b	141275.23b	786136.78ab	150063.82b	4505.75b
50 ppm	96119.67bc	71902.88c	650293.67ab	134352.42bc	3913.43bc
75 ppm	52769.91cd	46115.14c	301248.804b	55116.318c	3698.96bc
100 ppm	47016.92d	35111.37c	202322.57b	62833.13c	2654.30c
LSD at 0.05	45693.28	38077.12	1079396.20	86156.03	1301.680

The region of xylem cell in the *Gazania* was generally decreased with increase in salt contents (Fig.2). With the increase in salinity level there was noticeably reduction in xylem, phloem, cortex and epidermal area of leaf (Table 1).

Maximum xylem, phloem, cortex and epidermal region of leaf was observed in untreated plants while, minimum xylem, phloem, cortex, epidermal region of leaf was recorded in 50ppm, 75ppm and 100ppm solution of salt.

There was statistically no difference in leaf xylem and phloem region in those pots where levels of salinity were 25ppm and 50ppm applied. However there were statistically higher significant differences in 75ppm and 100ppm. Data pertaining to leaf lamina density indicates that with increasing in level of salinity there was significantly decreased in lamina thickness (Table 1).

Highest lamina density was founded in control while the average values of other treatments followed by 25ppm, 50ppm and 75ppm while, lowest lamina thickness was observed in plants with 100ppm salinity application. There was statistically highly noteworthy dissimilarity was observed in those plants where the salinity was not applied and where at 100ppm. It was also cleared that lamina density significantly decrease as compare to control (Fig.1). Whereas lower doses have lesser effect comparatively to higher concentrations on plants (Figs.2,3,4,5).

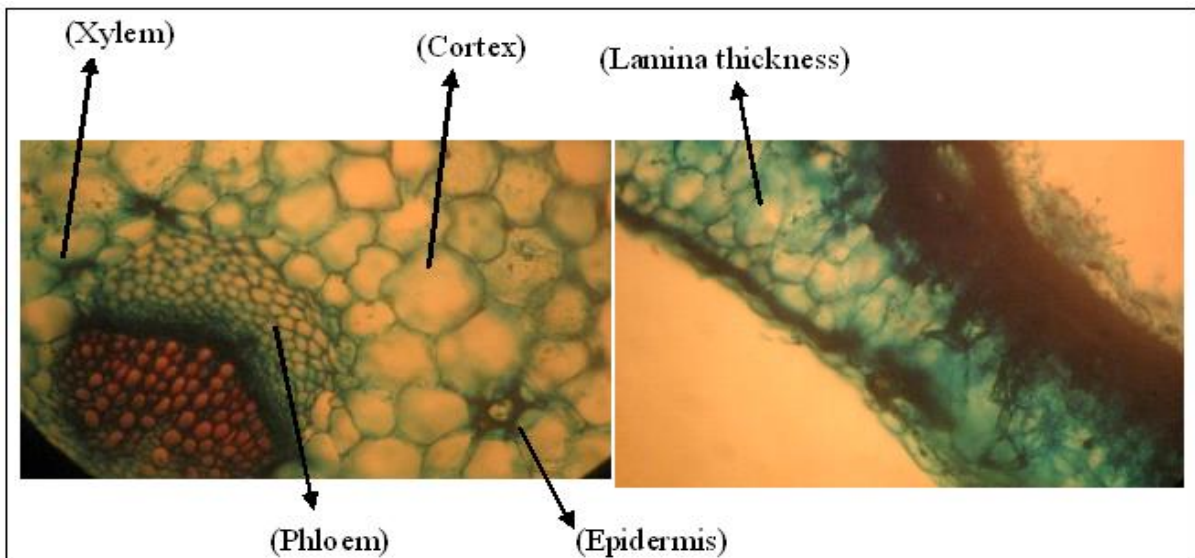


Fig. 1. Leaf anatomy of control plant.

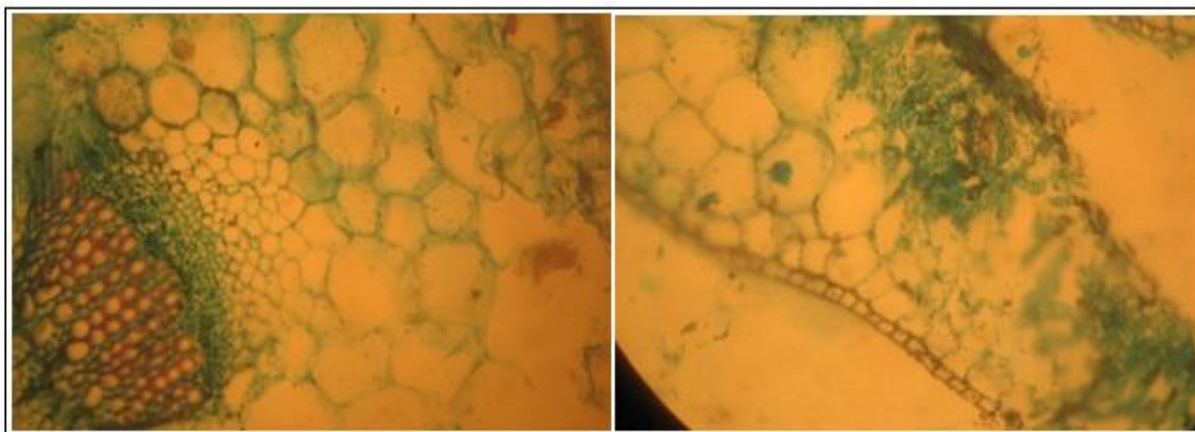


Fig. 2. Leaf anatomy of Gazania (25 ppm NaCl).

Discussion

In our experiment leaf anatomy was analyzed under different salinity situations. In plants under saline situations diff. anatomical variations take place (Huang and Van Steveninck, 1990; Winter, 1988).

These variations are different in each part of plant at diff. levels of development (Mills, 1989). Results in this study are clear that high salinity in soil has dissimilar effects on anatomy of the leaf as the region of xylem, phloem, cortex, lamina density, epidermis reduce in stress situation of salinity.

This caused by decrease of uptake of H_2O_2 by plants in high salinity situation and this lessening in H_2O_2 intake via cells formed osmotic form. Salinity cause reduction in phloem region and block that region at elevated levels, such conclusion were

interrelated with tolerant susceptible spp. (Goncharova and obrenkova, 1981). Salinity induced the steady variations in interior configuration of plants which minimizes the region of xylem and phloem (Hameed *et al.*, 2010).

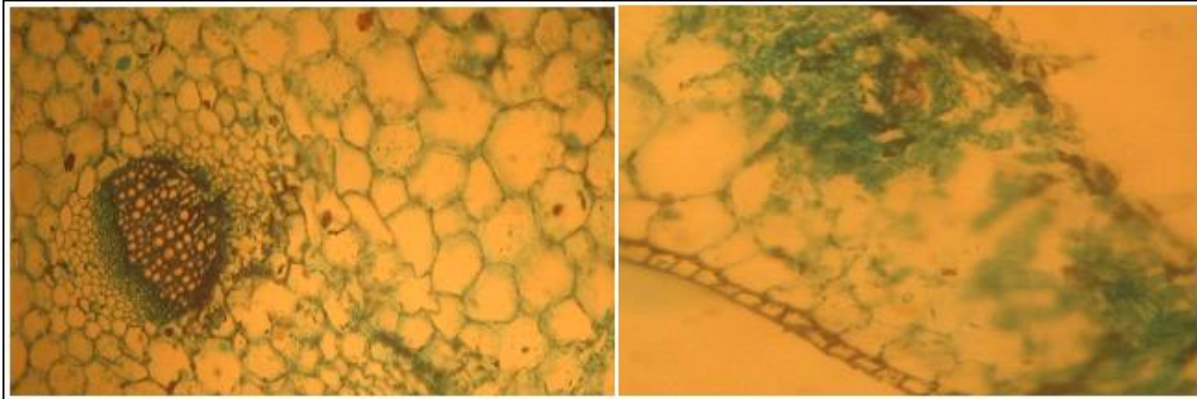


Fig. 3. Leaf anatomy of gazania (50 ppm NaCl).

These results are in accordance with findings of (Gadallah and Ramadan, 1997; Baum *et al.*, 2000; Gadalla, 2009) who reported that xylem area of leaf condensed due to reduction in xylem width and length. (Casenave *et al.*, 1999) observed that with amplification of salinity there was a reduction in development of xylem and cortex cotton seedlings. (Lingan *et al.*, 2002) reported that salinity stunted growth and caused variations in inner mechanism of

plant. Other similar results are in line with findings of (Hameed *et al.*, 2010) who observed that after a specific level of salinity noteworthy decline takes place in phloem area. Salt tolerant species are often characterized by thick inner tangential walls of endodermis, epidermis and lignified walls of cortical parenchyma, but at extreme level cortical thickness reduced (Baumeister and Merten, 1981; Hwang and Chen, 1995; Baloch *et al.*, 1998).

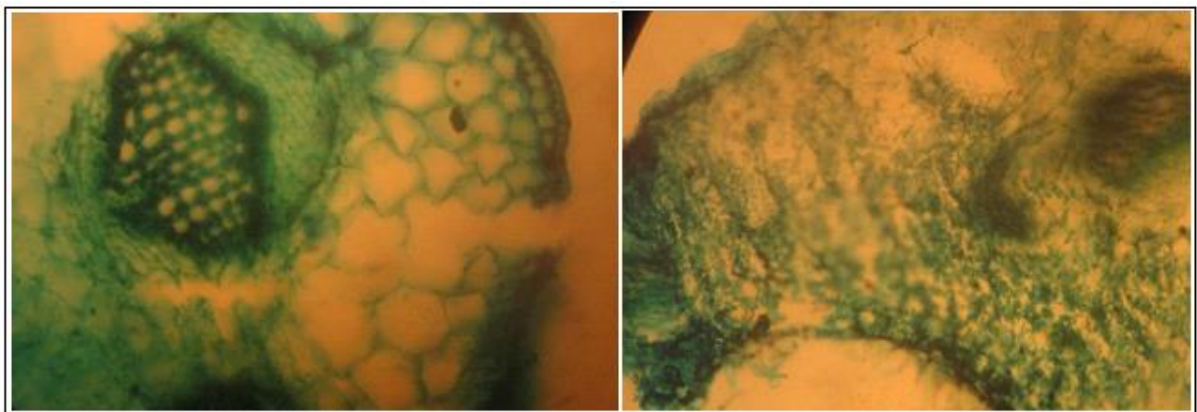


Fig. 4. Leaf anatomy of gazania (75 ppm NaCl).

Akram *et al.*, (2002) reported that cortical and pith region decreased in *Triticum aestivum* (L.) by reason of salinity, so acute parenchyma is a trait of salt tolerant spp. e.g salt grass (Alshammary *et al.*, 2004).

Similar results were suggested by (Curtis and Lauchli, 1987) who observed that salt stress condition reduced the epidermis cell width and length due to which area of epidermal cell also reduced.

This feature is crucial under partial humidity accessibility as density of epidermis region is able of inspection H₂O loss via stems (Hameed *et al.*, 2002; Nawazish *et al.*, 2006). Mansoor *et al.*, (2002) found that salinity reduces the development of epidermis due to changes in interior mechanism of plant.

Similar effect on leaf thickness has been reported previously for bean by (Meir, 1967) and (Wignarajah *et al.*, 1975) for *Phaseolus vulgaris* L. and for cotton by (Strogonov, 1964), as well as another species (Hayward, 1941; Poliakoff, 1975). (Nieman, 1962) observed that lamina thickness decreased under salt stress condition and suggested that salinity change the plant internal mechanism.

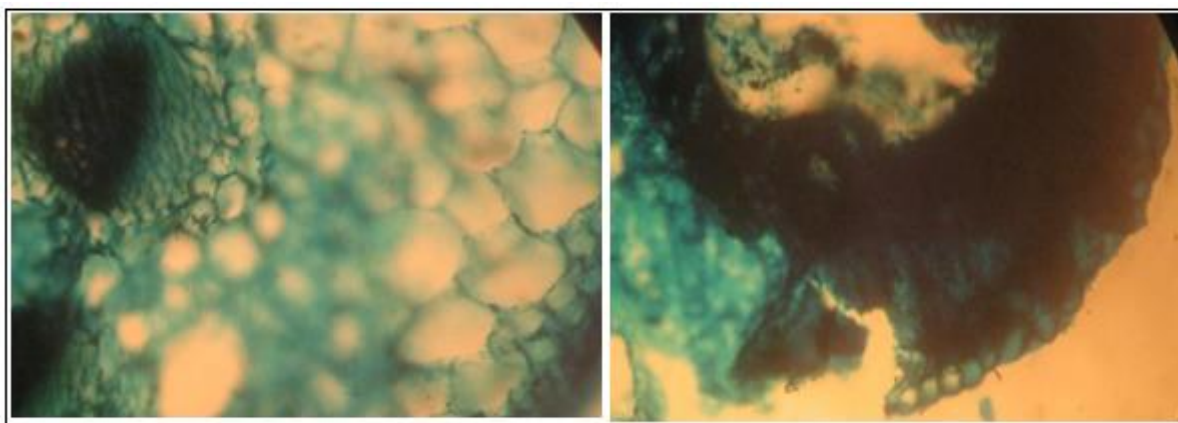


Fig. 5. Leaf anatomy of gazania (100 ppm NaCl).

Conclusion

From the above discussion, it is clear that stress situation due to salinity has crucial result t on overall pattern of plant development. Salinity has unenthusiastic relationship with morphological attributes. When we raise level of NaCl then it has unfavorable effect on anatomical characteristics of the plant. Between 5 levels of salinity, control exhibited good consequences beside other salinity treatments.

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