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EFFECT OF SOIL SALINITY ON LEAF DEVELOPMENT, STOMATAL SIZE AND ITS DISTRIBUTION IN COTTON (*GOSSYPIUM HIRSUTUM* L.)

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Abstract

Comparative effects of different salt levels (EC: 4-24 dS.m^{-1}) on the morphology and anatomy of four cotton cultivars viz., B-557, Niab-78, Sarmast and Qalandri was studied. A decrease in stomatal density under salt stress was compensated by an increase in stomatal size and mesophyll surface area. Adaptation to saline environment was adjusted by increasing mesophyll surface area to ensure normal exchange of gases and photosynthetic activities. In order of their salt tolerance, the cultivars studied can be rated as Niab-78 > B-557 > Qalandri > Sarmast.

Introduction

Salinity is known to effect seed germination, growth, reproduction with induced changes in anatomy and morphology of plants. A decline in total leaf area is often the first detectable response of salt or water stress in crop plants (Bradford & Hsiao, 1993). Ashley *et al.*, (1965) found a positive correlation between leaf area index and yield of cotton (*Gossypium hirsutum* L.) and exphasized that development of maximum foliage in the early growing season was essential for optimizing yield. Increase in leaf area was found to be more sensitive to salinity than either leaf emergence rate or dry matter accumulation in *Hibiscus cannabinus* (Curtis & Lauchli, 1987).

Leaves of cotton plants undergo anatomical and physiological modifications for ensuring better gas exchange capacity and are well adopted to grow under high winds, low humidity and frequent moisture stress. Gausman & Cardenas (1968) observed an increase in thickness of cotton leaves in plants growing at high salinity due to an excessive development of palisade and spongy mesophyll cells (Strogonov, 1962). Stomata exhibit variable resistance in the soil plant atmosphere continuum (Weatherley, 1976) thus controlling plant water status and gas exchange. These factors are also suggested to be genetically controlled. The present study reports the comparative effects of different salinity levels on morphological and anatomical characters of leaf, genetic potential of stomatal density and their size under non-saline and salt stress conditions in different cultivars of *G. hirsutum*.

Materials and Methods

Seeds of cvs. B-557, Niab-78, Sarmast and Qalandari, (all belonging to upland cotton, *Gossypium hirsutum*) were obtained from Cotton Research Station, Multan,

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Pakistan. Ten days old seedlings raised in normal soil at 3 leaf stage were transplanted in pots containing different concentrations of salinized soil. Plants were exposed to salt concentrations of 0.25, 0.5, 0.75 and 1% salinity (by weight) to give an EC_e value of 4, 16, 21, 22, and 24 $dS.m^{-1}$. The soil was made saline by mixing NaCl : $MgSO_4.7H_2O$: $CaCl_2.H_2O$: $NaHCO_3$ in the ratio of 9:8:2:1 (Abdullah *et al.*, 1978). Compost was mixed @ 600 g per 25Kg soil in each pot to improve the physical condition of soil. N,P and K were added @ 139 kg N, 50 Kg P and 25 Kg K per hectare (Huque, 1987).

Morphological & Anatomical Studies

Leaf Area: It was calculated by taking the square of the first right midrib x f as suggested by Johnson (1967).

Leaf Thickness: Free hand transverse sections were cut above the base of the fully matured leaf at the first sympodial node. Leaf thickness and area covered by spongy and palisade tissue were measured in ten leaf samples under the microscope.

Moisture Percentage: It was calculated as the percent difference between fresh and dry weight of samples.

Stomatal Counts: Mature leaves free 3rd/4th node below the apex were detached at dawn and dipped in Carnoy's solution to fix the tissue. Leaf samples were taken from middle where two central notches join by a line and observed under the microscope to record size and stomatal frequency/unit area.

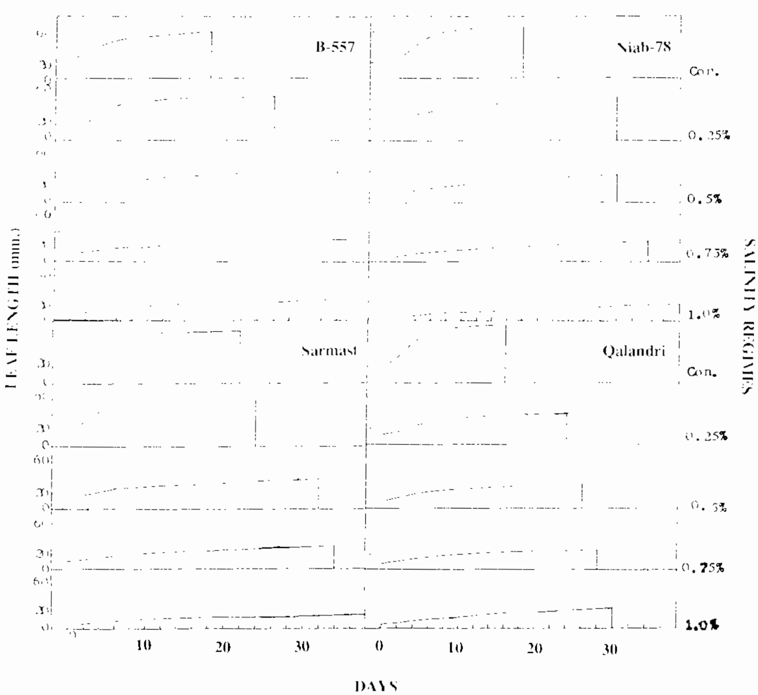

Fig. 1. Leaf development of different cotton cultivars as affected by different salinity regimes.

Relationship between mesophyll surface area and leaf surface area was quantified through a ratio A_{mes}/A in which A_{mes} is taken as total surface area of mesophyll and A as leaf surface area (Nobel, 1979).

Results and Dicussion

In crop plants, a decline in leaf expansion is often the first noticeable response to salt or water stress (Terry *et al.*, 1983). In the present study the development of leaf was completed in 18 days in cv. Qalandari followed by cvs. B-557, Niab-78 (20 days) and Sarmast (24 days) under control (Fig.1). Salt concentrations in the rooting medium significantly delayed leaf development in all cotton cultivars. Cv. Qalandari appeared most salt tolerant in which leaf development was completed in 32 days after its emergence which was followed by B-557 (33 days), Niab-78 and Sarmast (40 days) under 1% salinity regime (Fig.1). Similar delay in leaf development rate under saline conditions is reported in kenaf (Curtis & Lauchli, 1986).

Leaf area was also adversely effected by salt concentrations in the rooting medium and was significantly reduced by 66.04, 68.06, 79.08 and 83.81% in cvs. B-557, Qalandardi, Niab-78 and Sarmast in comparison with their respective control under 1% salinity regime (Fig.2). Reduction in leaf area of plants under salt stress has been reported in cotton (Hoffman *et al.*, 1971), potato (Abdullah & Ahmad, 1981), kenaf (Curtis & Lauchli, 1985) and maize (Kayani & Rehman, 1988).

The reduction in leaf area under salt stress is coupled with increase in leaf thickness as reported in cotton (Strogonov, 1962) and spinach (Robinson *et al.*, 1983. In the present studies, increase in leaf thickness was found to be 29.03% more as compared to control under 1% salinity regime in cv. B-557 followed by Qalandari (53.57%), Sarmast (56.25%) and Niab-78 (75.99%) (Table 1).

No significant differences were observed in moisture percentage of the leaves of the plants (Fig.3) grown under control and salinized condition indicating that increase in leaf thickness was due to accumulation of photosynthates. Increase in leaf thickness with corresponding increase in the ratio of palisade/spongy in cotton plants were also observed under different salinity regimes (Table 1). Such increase has been reported earlier in bean and cotton (Longstreth & Nobel, 1979).

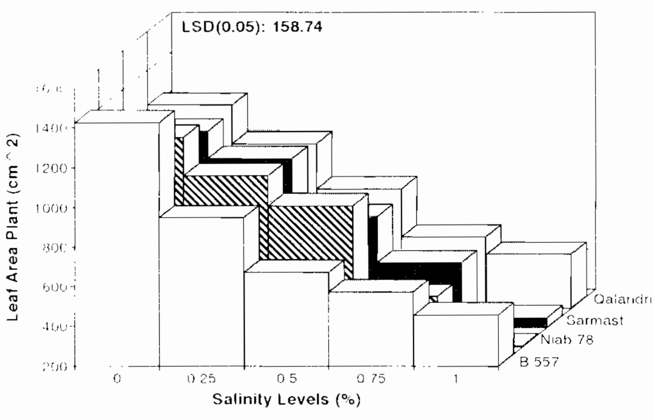

Fig.2. Total leaf area/plant of different cotton cultivars grown under different salinity regimes.

Table 1. Thickness and palisade/spongy ratio of leaves as affected by different salinity regimes in cotton cultivars.

	Salinity Treatments*									
Cultivars	Control	0.25%	0.5%	0.75%	1.0%	Control	0.25%	0.5%	0.75%	1.0%
	Leaf Thickness (μ)					Palisade/Spongy tissue ratio				
B-557	258.333 ±8.333	275.000 ±15.000	283.333 ±8.333	308.333 ±20.321	333.333 ±20.047	0.804 ±0.105	0.883 ±0.072	1.250 ±0.000	0.952 ±0.041	1.108 ±0.256
Niab-78	208.333 ±8.333	283.333 ±16.047	304.166 ±4.166	316.666 ±16.047	366.666 ±20.046	0.916 ±0.083	0.923 ±0.189	0.938 ±0.115	1.148 ±0.148	1.158 ±0.163
Sarmast	200.000 ±14.433	225.000 ±14.433	241.666 ±16.666	270.833 ±4.166	312.500 ±19.094	0.938 ±0.001	1.212 ±0.038	1.109 ±0.000	1.209 ±0.012	1.163 ±0.010
Qalandari	233.333 ±12.047	237.500 ±12.500	266.666 ±12.000	308.333 ±12.047	358.333 ±16.324	1.000 ±0.000	0.915 ±0.004	1.196 ±0.023	1.257 ±0.028	1.240 ±0.029

*Salinity treatments refers to the addition of salt mixture as percentage in soil.

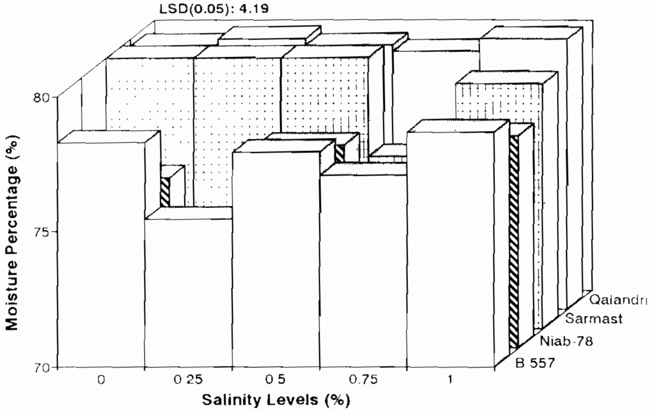

Fig.3. Moisture percentage in leaves of cotton cultivars grown under different salinity regimes.

There was an increase in stomatal frequency/unit area of leaf under saline conditions in all the cotton cultivars (Table 2). A reduction in the total number of stomata leaf started from 0.75% salinity regime. Such reduction under salt stresses has been reported in cotton plants (Stroganov, 1962) and in *Hibiscus cannabium* (Curtis & Lauchli, 1985). Decrease in the number of stomata under stressed condition associated with partial closing of aperture upsets gaseous exchange thus distrurbing normal photosynthetic process. Gale *et al.*, (1967) have reported reduction in net photosynthesis in onion, bean and cotton due to decrease in diffusion of CO_2 as a result of partial stomatal closure.

Plants growing under salt stress develop certain adaptive modifications in leaf structure to ensure regular gaseous exchange. Present studies indicate that reduction in the number of stomata under high salinity is partly compensated by an increase in the size of guard cells (probably due to greater succulence) which could improve efficiency of gasous exchange (Table 2).

Since salinity effects have modified the stomatal adaptation in all *G. hirsutum* cultivars under saline environment with non-significant difference, the increased mesophyll surface area is considered as the criteria of salt tolerance. It carries beneficial effects on gaseous exchange and to carry out photosynthesis process. On the basis of increase mesophyll surface area, the cultivars can be rated in the following order of salt tolerance, Niab-78 > B-557> Qalandari > Sarmast.

Table 2. Stomatal density and morphology of cotton cultivars as affected by different salinity regimes.

Salinity Treatments*	Stomatal frequency/unit area				Stomatal density/leaf (in 000)				Stomatal size (μ)			
	B557	Niab78	Sarmast	Qalandari	B557	Niab78	Sarmast	Qalandari	B557	Niab78	Sarmast	Qalandari
Control	13.0	10.66	10.66	13.33	76.00	54.95	56.88	78.88	26.943	26.944	24.721	26.944
	±2.0	±0.33	±1.66	±2.60	±17.81	±2.61	±9.29	±18.53	±1.460	±1.944	±2.221	±1.001
0.25%	15.66	13.00	11.33	13.66	80.44	60.12	61.19	66.59	27.221	27.220	26.249	28.034
	±2.72	±3.05	±2.02	±2.66	±14.53	±5.88	±8.67	±14.67	±0.735	±0.555	±0.867	±1.211
0.5%	19.00	13.33	12.00	14.66	81.92	65.59	60.77	73.94	28.888	28.054	26.944	28.609
	±2.00	±1.33	±1.52	±3.75	±11.82	±8.78	±9.19	±19.29	±0.555	±0.734	±1.001	±1.21
0.75%	15.00	13.33	14.66	16.00	67.10	51.93	54.52	66.37	29.110	28.296	27.221	28.888
	±1.73	±2.02	±1.20	±2.08	±10.08	±17.61	±6.24	±21.24	±0.484	±0.963	±1.389	±1.944
1.0%	12.00	14.00	16.00	16.33	41.53	39.44	33.45	48.54	31.943	29.165	27.499	29.721
	±0.57	±2.08	±0.57	±2.72	±12.31	±9.60	±7.64	±8.47	±1.821	±1.273	±0.481	±0.278

*Salinity treatments refers to the addition of salt mixture as percentage in soil.

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