The effects of salt stress on growth physiology of selected okra (*Abelmoschusesculentus* L.) Cultivars

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Abstract

Salinity due to over-accumulation of NaCl is usually of great concern and it is the most injurious factor in arid and semi arid regions. Considering this feature, an experiment was conducted to evaluate the salinity stress responses of selected okra cultivators on Diffusive Resistance (DR) and Relative Water Content (RWC). The okra cultivars 'Haritha', 'EUOK2' and 'MI5' were used for this study. Sodium chloride (100mM) was used to create the salinity while, distilled water was used as control. Salt stress significantly increased the DR of the selected okra cultivars. The highest increase (5.1 scm^{-1})was obtained in the 'MI5' followed by 'Haritha'(4.2scm^{-1}) and 'EUOK2'(3.6 scm^{-1}). The highest increase found in the 'MI5' okra cultivar would have been due to tight closure of stomata in this cultivar compared to the others. The lowest value found in the 'EUOK2' would have been due to its inherent characteristic feature. The stomates of this cultivar would have kept opened relatively widely than the others. Salt stress significantly reduced the RWC of the selected okra cultivars. The highest reduction (58.7 %) was found in the'MI5' followed by 'Haritha' (67.3 %) and 'EUOK2' (73.5 %). The 'EUOK 2' maintained relatively high amount of water in their leaf tissues under salinity stress. All these events indicate that 'EUOK 2' cultivar of okra had the ability to withstand salinity stress much better than the others. As a result, 'EUOK2' was recognized as the most salinity tolerant okra cultivar which could be grown in the salt affected areas of the sandy regosols.

Keywords: Diffusive resistance, Okra, Relative water content, Salinity stress.

Introduction

Among a biotic stresses, salinity is one of the most severe problems in worldwide agricultural production. Salinity is defined as the amassment of water-soluble salts in the top layer of soil to a level that drastically affects crop production (Rengasamy, 2002). Salinization of soils is one of the serious problems for irrigated agriculture, and the situation is most severe in tropical regions (Khan *et al*., 2003). High salt contents reduce the growth and production by affecting physiological processes, including modification of ion balance, water status, mineral nutrition, stomatal behavior and photosynthetic efficiency (Munns,1993).

Most plants are salt sensitive with either a relatively low salt tolerance or severely inhibited growth at low salinity levels so differ in the growth response to salinity (Moisender*et al*., 2002). High concentration of salts in the root zone decreases soil water potential and the availability of water (Lloyd *et al*., 1989). This deficiency in available water under saline condition causes dehydration at cellular level and ultimately osmotic stress occurs. The higher ratios of toxic salts in leaf apoplasm lead to dehydration and turgor loss and death of leaf cells and tissues (Marschner, 1995).

The ion toxicity has negative impacts on plant growth and development due to low water potential within the local root environment. Ions such as sodium (Na^+) and chloride (Cl^-) (>40 mmol/L) can be toxic to plants at levels that result in an imbalance in plant nutrition due to decreased nutrient uptake and transport to new shoots (Munns and Tester, 2008). High salinity modifies plant metabolisms, which results in altered plant morphology; cultivar type, duration, and intensity of stress determine the extent of morphological modification (Khan *et al*., 2003). The excess salt can cause decreased seed germination, seedling growth, and dry matter production (Janila *et al*., 1999) also induces Fe^{2+}, K^+, and Ca^{2+} deficiencies, resulting in yield losses (Singh *et al*., 2004).

Okra (*Abelmoschusesculentus L.)* is recognized as an annual herbaceous plant grown in tropical and subtropical areas and serves as a source of carbohydrates, fats, vitamins and various minerals (Oyenuga, 1968). In spite of having good nutritional value, its per hectare yield is very low. Okra cultivation is hampered in saline and sodic (having more Na^+) soil as it is sensitive to salinity (Ashraf *et al.,* 2003).This decline in optimum yield is due to the drastic effects of salts, which are deposited in soil by the use of brackish underground water and addition of industrial effluents in canals also adds salts in irrigation water. Okra plant at earlier growth stages is more sensitive to salinity (Cedra*et al*., 1982). While later on, the ionic stress in turn reduces leaf expansion. During long term exposure to salinity, plants experience ionic stress, which can lead to premature senescence of adult leaves and thus reduction in photosynthetic rate is a common observation (Cramer and Nowak, 1992). Physiological mechanisms like ion exclusion, ion accumulation, production of compatible solutes, and osmotic adjustment are considered to be associated with varietal characteristics in relation to salt tolerance.

The present study was conducted to determine if the diffusive resistance and relative water content measurements on okra plants could be used to differentiate between cultivars differing in apparent salt tolerance, to deal with the physiological responses manifested as a result of salinity stress and to suggest the most salt tolerant okra cultivar that could be grown under salinity situation.

Materials and Methods

This experiment was conducted at the Agronomy farm of the Eastern University which is located at an elevation of 75m above mean sea level in the Eastern Province of Sri Lanka. Studies were conducted during the '*Yala*' season of the year 2014. The climate is warm with the temperature of 28 - 32°C and an average annual rain fall of 1250 mm.

Okra (*Abelmoschusesculentus* L.) cvs. 'Haritha', 'EUOK 2' and 'MI5' were used for this study. The seeds were surface sterilized with sodium hypochlorite 0.5% (v/v) for 20 min. washed repeatedly with distilled water and were allowed to germinate in polyethylene bags filled with fine sand as growth medium. A number of three seeds per bag were sown initially but, after 15 days of germination, the plants were thinned out to one.Plants were grown in Hoagland (Hoagland and Arnon, 1950) solution under non saline conditions for 30 days after germination. Afterwards, the salt treatment was imposed. Sodium chloride was dissolved in distilled water to obtain the concentration of 0 (control) and 100mM(Normal saline condition) and this solution was applied to create the salinity while half strength Hoagland solution was applied as nutrient medium. The treated plants were grown under saline condition. Irrigation along with half strength Hoagland solution was applied to the selected treatments according to the need of the plants by regularly observing the moisture content of sand.

The experiment was carried out with six treatments and five replications and the treatments were as follows:

T_1 = 'Haritha' cultivar of okra irrigated with distilled water (Control)

T_2 = 'Haritha' cultivar of okra irrigated with saline water (100mMNaCl)

T_3= 'EUOK2' cultivar of okra irrigated with distilled water (Control)

T_4= 'EUOK2' cultivar of okra irrigated with saline water (100mMNaCl)

T_5= 'MI 5' cultivar of okra irrigated with distilled water (Control)

T_6= 'MI 5' cultivar of okra irrigated with saline water (100mMNaCl)

The experiment was laid out in a Completely Randomized Design with 2 x 3factor Factorial arrangements.

Physiological Attributes

Diffusive Resistance (DR)

A number of six leaves representing six plants were randomly selected from each replicate of the treatments for the measurement of DR. This parameter was measured by a Portable Steady State Porometer (LI-1600, LI-COR Inc. USA) on the 15^{th} day of salinity application for the treated and control plants. This measurement was made between 9-11 am when the Photosynthetically Active Radiation (PAR) was above the saturation PAR of 1500 $\mu ES^{-1} M^{-1}$.

Relative Water Content (RWC)

A number of six leaves representing six plants were randomly collected from each replicate of the treatments for the determination of RWC. Similar sized discs (3 cm diameter) were obtained from the leaves using a cork borer and their fresh weights (FW) were recorded soon after collection. These discs were placed overnight in a beaker containing deionized water to obtain turgid weight (TW). The beaker containing water was placed inside the refrigerator to avoid physiological deterioration of leaf cells. The leaf discs were blotted with filter papers to remove the excess water and their TW was recorded. The leaf discs were then placed in the oven at 80°C for 24 hours and their dry weights (DW) were recorded.

$$\text{Relative Water Content } (\%) = \frac{(\text{Fresh Weight} - \text{Dry Weight})}{(\text{Turgid Weight} - \text{Dry Weight})} \text{ X } 100$$

The data were analyzed through Statistical Analysis System (SAS) software statistical package and the differences between means were compared using DMRT.

Results and Discussion

It was found that there were significant differences between treatments in the Diffusive Resistance (DR) and Relative Water Content (RWC) of leaves of the selected okra cultivars.

Diffusive Resistance (DR)

In the treatments where the salinity stress was imposed on plants, the DR on the 15th day of salinity application was significantly higher than the control values (Table 1).

Table 1: The effects of salinity stress on the Diffusive Resistance (DR) of selected okra cultivars

Cultivars	**Diffusive Resistance (scm^{-1})**	
	Control	**Salinity stress**
'Haritha'	(T_1) 1.2 ^{b}	(T_2) 4.2 ^{a}
'EUOK 2'	(T_3) 0.9 ^{b}	(T_4) 3.6 ^{c}
'MI 5'	(T_5) 1.1 ^{b}	(T_6) 5.1 ^{d}

*Values in the same column followed by the same letter do not differ significantly (P< 0.05)
*Values are the means of 30 plants in 5 replications.

It was also found that there were significant differences in the DR values of salinity stressed okra cultivars. The highest DR was obtained in the 'MI 5' cultivar and the lowest was found in the 'EUOK 2'. 'Haritha' showed the DR value in between the above two cultivars. Exposure of okra plants to salinity stress increased the diffusive resistance of these plants. As pointed out by Chatrath*et al*. (2000), salinity increased the stomatal resistance, which could be explained by inhibition of plant growth to water stress. Turan*et al.* (2007) stated that there was a strong negative correlation between stomatal resistance and NaCl. In general, measurement of stomatal resistance provides effectual comparison for determining the degree of salt stress in plants. During a salt stress, the plant has to close their stomata due to water loss. Stomatal factors have also a more significant effect on photosynthesis (Wang *et al*., 1987).

The lowest DR value observed in the 'EUOK2' under saline condition indicates that the stomates of this cultivar would have opened comparatively wider than the stomates of the other two cultivars. As a result, 'EUOK2' cultivar would have had better gas exchange capacity than the rest of the cultivars. Perhaps, this would have increased the photosynthetic rate of this cultivar compared to the others. The highest DR value observed in the 'MI 5' indicates that the stomates of this cultivar would have closed much more tightly than those of 'EUOK2' and 'Haritha' cultivars. As a consequence, reduced gaseous exchange followed by reduction in photosynthetic rate would have occurred. Based on these observations it could be stated that the 'EUOK 2' cultivar of okra exhibited better salt tolerance than the other two cultivars. 'MI 5' showed salt susceptibility among the tested cultivars.

Relative Water Content (RWC)

In the treatments where the salinity stress was imposed on plants, the RWC on the 15th day of salinity application was significantly lower than the control values.It was found that there were significant differences in the RWC values of okra cultivars which were exposed to salinity stress (Table 2).

Table 2: The effects of salinity stress on the Relative Water Content (RWC) of selected okra cultivars

Cultivars	**Relative Water Content (%)**	
	Control	**Salinity stress**

'Haritha'	(T_1) 87.6 [a]	(T_2) 67.3 [b]
'EUOK 2'	(T_3) 84.1 [a]	(T_4) 73.5 [c]
'MI 5'	(T_5) 86.3 a	(T_6) 58.7 [d]

*Values in the same column followed by the same letter do not differ significantly ($P< 0.05$)
*Values are the means of 30 plants in 5 replications.

The highest RWC was obtained in the 'EUOK 2' whereas the lowest was found in the 'MI 5'. The RWC of 'Haritha' cultivar was in between these two cultivars. Salinity stress reduced the RWC of all the okra cultivars. Appraisal of water relations in plants grown under stress conditions including saline stress is necessary to ascertain that up to what extent cellular water content is maintained, because almost all metabolic activities within the cell are dependent on the availability of sufficient amount of water therein (Ashraf *et al*,. 2011).As stated by Shaheen*et al*. (2013), salt stress significantly reduced the relative water content. Based on this observation it could be stated that the 'EUOK 2' cultivar was able to maintain relatively high RWC than the other two cultivars under salinity condition. This is a favorable feature with regard to salt tolerance of this cultivar. Cultivars which were believed to be more salt resistant usually maintain higher leaf RWC under salinity stress. The lowest RWC observed in the 'MI 5' cultivar exhibits its susceptibility to salt stress.

Conclusions

Soil salinity, one of the most serious problems on planting areas, has the most obstructive impact on crop production in the world. This salinity problem attracts many scientists to overcome this obstruction by improving salt tolerant cultivars. These results indicated that all the tested cultivars exhibited reduction in physiological attributes as a result of an increase in salinity level. The reduction in growth physiology of okra may be due to the inhibitory effect of the accumulated ions of sodium and chloride on the metabolic activities. This study determined the extent to what the Diffusive Resistance and Relative Water Content of selected okra cultivars were impaired by salt stress during the crop growth period. The responses were exhibited. Okra cultivar 'EUOK 2' was able to resist salt stress relatively better than the other two cultivars. It could therefore be concluded that 'EUOK 2' okra cultivar is the most salt tolerant one among the tested okra cultivars which could thrive in the salt affected areas of the sandy regosols.

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