Anales de Biología 30: 17-28, 2008

Growth, water status and nutrient accumulation of seedlings of *Acacia senegal* (L.) Willd. in response to soil salinity

Seema Abhay Hardikar[1] & Amar Nath Pandey[2]

1 Department of Biosciences, Saurashtra University, Rajkot, 360005 India, e-mail:abhay.hardikar@sgs.com; seemahardikar@yahoo.com

2 Department of Biosciences, Saurashtra University, Rajkot – 360005 India, e-mail:anpandey2001@gmail.com

Correspondence
A. N. Pandey
Email: anpandey2001@gmail.com
Tel (O): +91-281-2586419
Tel (M): +919427495989
Fax (O): +91-281-2577633
Received: 2 February 2008.
Accepted: 30 July 2008.

Resumen

Crecimiento, estado hídrico, y acumulación de nutrientes en plántulas de Acacia senegal *(L.) Wild. en respuesta a la salinidad del suelo.*

Se llevaron a cabo una serie de experimentos en invernadero con el fin de evaluar los efectos de la salinidad del suelo sobre la emergencia, crecimiento, estado hídrico, contenido de prolina y acumulación de minerales de plántulas de *Acacia senegal* (L.) Willd.(Mimosaceae). Se añadió NaCl al suelo y se mantuvo la salinidad a 0.2, 3.9, 6.2, 8.1, 10.0 y 11.9 dSm^{-1}. La salinidad causó reducción del contenido de agua y del potencial hídrico de los tejidos, lo que resultó en un déficit interno de la planta. Consecuentemente, el crecimiento de las plántulas disminuyó significativamente, mientras que el contenido de prolina de los tejidos aumentó. No aparecieron mecanismos efectivos para controlar la absorción de Na y su transporte a los tejidos de los brotes. El contenido de N, P, K y Ca disminuyó significativamente en los tejidos como respuesta a la salinidad. Se discute sobre los cambios en los tejidos y el patrón global de acumulación de otros elementos, así como posibles mecanismos para evitar la toxicidad del Na en esta especie arbórea.

Palabras clave: Salinidad del suelo, Crecimiento de plántula, Contenido en prolina, Potencial hídrico, Macro/micronutrientes, Tolerancia a la sal.

Abstract

Greenhouse experiments were conducted to assess the effects of soil salinity on emergence, growth, water status, proline content and mineral accumulation of seedlings of *Acacia senegal* (L.) Willd. (Mimosaceae). NaCl was added to the soil and salinity was maintained at 0.2, 3.9, 6.2, 8.1, 10.0 and 11.9 dSm^{-1}. Salinity caused reduction in water content and water potential of tissues, which resulted in internal water deficit to plants. Consequently, seedling growth significantly decreased with increase in soil salinity. Proline content in tissues increased with increase in soil salinity. There were no effective mechanisms to control net uptake of Na and its transport to shoot tissues. N, P, K and Ca content significantly decreased in tissues in response to salinity. Changes in tissues and whole-plant accumulation patterns of other elements, as well as possible mechanisms to avoid Na toxicity in this tree species in response to salinity, are discussed.

Key words: Soil salinity, Seedling growth, Proline content, Water potential, Macro- and micro-nutrients, Salt tolerance.

Introduction

Saline soils are abundant in semi arid and arid regions where the amount of rainfall is insufficient for substantial leaching (Marschner 1995). Salinity is a scourge for agriculture, forestry, pasture development and other similar practices. An understanding of responses of plants to salinity is of great practical significance. High concentrations of salts have detrimental effects on plant growth (Taiz & Zeiger 2006, Ramoliya et al. 2006) and excessive concentrations kill growing plants (Garg & Gupta 1997). There occurs retardation of germination and growth of seedlings at high salinity (Bernstein 1962, Garg & Gupta 1997, Ramoliya et al. 2006). However, plant species differ in their sensitivity or tolerance to salts (Marschner 1995). There are evidences that organs, tissues and cells at different developmental stages of plants exhibit varying degrees of tolerance to environmental conditions (Munns 1993). It is reported that soil salinity suppresses shoot growth more than the root growth (Maas & Hoffman 1977, Munns 2002, Ramoliya et al. 2006).However, fewer studies on the effect of soil salinity on root growth have been conducted (Munns 2002). The high salt content lowers osmotic potential of soil water and consequently the availability of soil water to plants. The salt–induced water deficit is one of the major constraints for plant growth in saline soils. In addition, many nutrient interactions in salt–stressed plants can occur that may have important consequences for growth. Internal concentrations of major nutrients and their uptake have been frequently studied (e.g. Cramer et al. 1989, Maas & Grieve 1987, Ramoliya et al. 2006, Patel & Pandey 2007), but the relationship between micro-nutrient concentrations and soil salinity is rather complex and remains poorly understood (Tozlu et al. 2000). An understanding of growth and survival of plants under saline habitat conditions is needed for (i) screening the plant species for the afforestation of saline deserts and (ii) understanding the mechanism that plants use in the avoidance and /or tolerance of salt stress.

Acacia senegal (L.) Willd. (Mimosaceae), a small deciduous tree species grows abundantly in coastal forests of Saurashtra in Gujarat State of India. It also grows successfully on marginal-saline lands of Kutch (north-west saline desert) contiguous to Saurashtra. *A. senegal*, yields commercial gum arabic. Wood is a good fuel. Leaves and pods are eaten by herbivores. The present investigation was performed with the following objectives: (i) to understand the adaptive features of *A. senegal* which allow it to grow and survive in saline and arid regions and (ii) to assess the pattern of macro– and micro–nutrient accumulation within the tissues of this tree species in response to salt stress.

Material and Methods

Study area

The present study was carried out in a greenhouse of the botanical garden of Saurashtra University at Rajkot (22º 18' N Lat, 70º56' E Long) in Gujarat. For the emergence and growth of seedlings the top 15 cm black-cotton soil which is predominant in Saurashtra region of Gujarat, was collected from an agricultural field. This soil is a clayey loam containing 19.6% sand, 20.3% silt and 60.1% clay. The available soil water between wilting coefficient and field capacity ranged from 18.3% to 35.0%, respectively. The total organic carbon content was 1.3% and pH was 7.2. The electrical conductivity of soil was 0.2dSm^{-1}, Nitrogen, phosphorus, potassium, calcium and sodium contents were 0.15%, 0.05%, 0.03%, 0.05% and 0.002%, respectively. This soil is fertile and fit for intensive agriculture. Physical and chemical properties of soil are given earlier (Patel & Pandey 2007). The Kutch and Saurashtra regions are tropical monsoonic and can be ecoclimatically classified as arid and semi-arid, respectively. The entire area is markedly affected by south-western monsoon which causes the onset of wet season in mid – June, and its retreat by the end of September coincides with a lowering of temperature and gradual onset of winter. Total annual rainfall is about 395mm at Bhuj (23º15' N Lat, 69º49' E Long) in Kutch and about 554mm at Rajkot in central Saurashtra which occurs totally during the rainy season. Typically, there are three main seasons: summer (April - mid June), monsoon (mid June–September) and winter (November–February).

The months of October and March are transition periods between rainy (monsoon) and winter and between winter and summer seasons, respectively. Winters are generally mild and summers are hot.

Salinisation of soil

Surface soil was collected, air dried and passed through a 2 mm. mesh screen. Six lots of soil of 100 kg each, were separately spread over about 50 mm thick polyethylene sheets. Sodium chloride (NaCl) amounting to 390, 700, 1070, 1275 and 1530g was then thoroughly mixed with soil of five lots, respectively to give electrical conductivities of 3.9, 6.2, 8.1, 10.0 and 11.9 dSm^{-1}. There was no addition of NaCl to sixth lot of soil that served as control. The electrical conductivity of control soil was 0.2dSm^{-1} and this value was approximately equal to 2 mM salinity. For the measurement of electrical conductivity a soil suspension was prepared in distilled water at a ratio of 1:2 in terms of weight. The suspension was shaken and allowed to stand overnight. Thereafter, electrical conductivity of the supernatant solution was determined with a conductivity meter.

Seedling emergence

Twenty polyethylene bags for each level of soil salinity were each filled with 5 kg of soil. Tap water was added to each bag to bring the soils to field capacity and soils were allowed to dry for 7 days. Soils were then raked using fingers and seeds were sown on 18 August 2006. Seeds of *A. senegal* were collected from the coastal area of Arabian sea in Jamnagar city of Saurashtra. Bags were kept in a greenhouse. Ten seeds were sown in each bag at a depth of 8-12 mm. Immediately after sowing soils were watered and thereafter watering was carried out on alternate days. Emergence of seedlings was recorded daily over a period of 40 days. A linear model was fitted to cumulative proportion of seed germination and increasing soil salinity, using the expression:

$$Sin^{-1}\sqrt{p} = \beta_0 + \beta_1 X$$

where, $Sin^{-1}\sqrt{p}$ is cumulative proportion of seed germination, X is soil salinity and β_0 and β_1 are constants. Salt concentration at which seed germination was reduced to 50% (SG_{50}) was estimated using the model.

Seedling growth

For the growth studies, two seedlings that emerged first were left in each of 20 bags at each level of salinity and others were uprooted. Seedlings grown in soils at 0.2, 3.9 and 6.2 dSm^{-1} salinity exhibited emergence of the second leaf after 15 days whereas the second leaf on seedlings grown in 8.1, 10.0 and 11.9 dSm^{-1} appeared after 23 days. Emergence of the second leaf confirmed the establishment of seedlings. However, only 19.6% seed germination was recorded in soil at 11.9 dSm^{-1} salinity, and further experiments were not conducted on those seedlings. Following emergence of the second leaf, one seedling having better vigor was allowed to grow in each bag and another seedling was further uprooted. Thus twenty replicates factorialized with five grades of soil (0.2, 3.9, 6.2, 8.1, and 10.0dSm^{-1}) were prepared. This gave a total of 100 bags, which were arranged in 20 randomized blocks. Seedlings were watered (about 250 ml water was added to raise the soil moisture to field capacity) on alternate days and allowed to grow for six months. Experiment was terminated on 18 February 2007. Five plants grown in soil at 10 dSm^{-1} salinity died during the course of experiment. Seedlings contained in 15 bags at each salinity level were washed to remove soil particles adhered with roots. Morphological characteristics of each seedling were recorded. Shoot height and root length (tap root) were measured. Leaf area was marked out on graph paper. Fresh and dry weights of leaves, stems, tap roots and lateral roots were determined. Sum of leaf and stem weight was considered as shoot weight. Water content (gg^{-1} dry weight) in plant tissues (leaves, stems, tap roots and lateral roots) was calculated using fresh and dry weight values. Data recorded for morphological characteristics, dry weight and water content of different components were analyzed by one way ANOVA to assess the effect of salinity on plant growth.

Determination of water potential and proline content

Five additional plants grown in soil at each level of salinity were used for measurement of water potential and proline estimation in plant tissues. Water potential of leaves, stems, tap roots and lateral root tissues was measured by Dewpoint Potential Meter WP4. Concentration of proline in plant tissues was estimated following Bates et al. (1973). Extract of 0.5g fresh plant material with aqueous sulphosalicylic acid was prepared. The extracted proline was made to react with ninhydrin to form chromophore and read at 520 nm. Water potential and proline content of tissues were estimated in triplicate. Data were analyzed by one way ANOVA.

Mineral analyses of plant materials

Mineral analyses were performed on leaves, stems, tap roots and lateral root tissues. Plant parts of the seedlings grown in soil at same level of salinity were pooled separately. Plant samples were ground using mortar and pestle. Three sub samples of plant tissues were analyzed. Total nitrogen was determined by Kjeldahl method and phosphorus content estimated by the chlorostannous molybdophosphoric blue colour method in sulphuric acid (Piper 1944). Concentrations of Ca, Mg, Na, K, Zn, Fe, Mn and Cu were determined by Shimadzu double beam atomic absorption spectrophotometer AA-6800 after triacid (HNO_3: H_2SO_4:$HClO_4$ in the ratio of 10:1:4) digestion. Mineral data were analyzed by one way ANOVA. Correlations and linear regression equations between mineral content and salt concentrations were determined.

Results

Effect of salinisation on seedling emergence

Seedlings began to emerge 3 days after sowing and 84.8% seed germination was obtained over a period of 20 days, under control (0.2 dSm^{-1} salinity) conditions (Fig.1). Seedling emergence in saline soils was also recorded 3 days after sowing. Seedling emergence lasted for 18, 19, 18, and 18 and 12 days in soils with 3.9, 6.2, 8.1, 10.0 and 11.9 dSm^{-1} salinities, respectively and corresponding seed germination was 62.8%, 58.4%, 50.4%, 44% and 19.6%. There was a significant reduction in seed germination ($p<0.01$) with increasing salt stress. A negative relationship between proportion of cumulative seed germination and concentration of salt was obtained according to the following expression:

$Y= 68.654 - 3.134\ X$, (R^2_{adj} =0.937, $p<0.01$)

where Y is arcsine (degrees) of proportion of cumulative seed germination and X is salt concentration.Increasing soil salinity significantly retarded ($p<0.01$) stem and root elongation (Fig. 1). Nevertheless, root length was remarkably greater than shoot height for seedlings grown in control and saline soils. There was a negative relationship ($p<0.01$) for shoot height and root length with increase in salt concentration in soil. Leaf expansion was significantly reduced ($p<0.01$) by increasing concentration of salt in soil. A negative relationhip was obtained between leaf area and salt concentration ($p<0.01$).

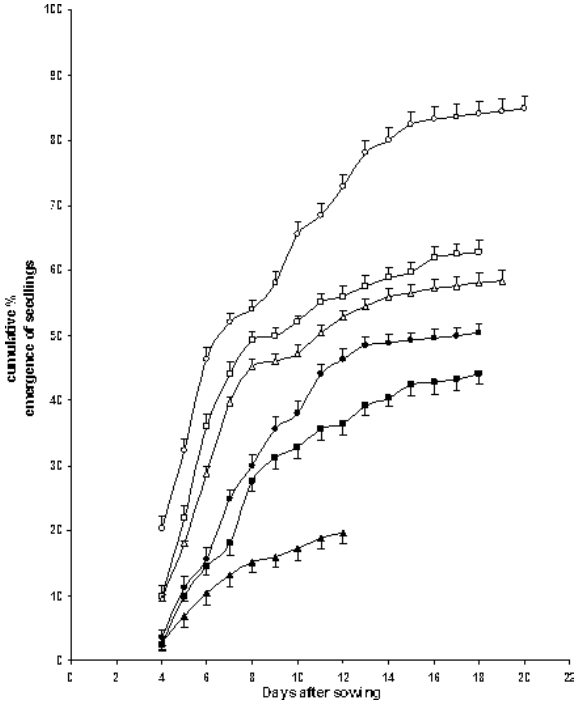

Figura 1. Emergencia acumulada de plántulas de *Acacia senegal* en respuesta a la salinidad del suelo. 0.2dSm-1(○), 3.9dSm-1(□), 6.2dSm-1(Δ), 8.1dSm-1(●), 10.0dSm-1(■) y 11.9dSm-1(▲). Las barras de error representan el SE.

Figure 1. Cumulative emergence of seedlings of *Acacia senegal* in response to soil salinity. 0.2dSm-1(○), 3.9dSm-1(□), 6.2dSm-1(Δ), 8.1dSm-1(●), 10.0dSm-1(■) and 11.9dSm-1(▲). Error bars represent SE.

Effect of salinisation on stem and root elongation and leaf expansion

Increasing soil salinity significantly retarded ($p<0.01$) stem and root elongation (Fig. 1). Nevertheless, root length was remarkably greater than shoot height for seedlings grown in control and saline soils. There was a negative relationship ($p<0.01$) for shoot height and root length with increase in salt concentration in soil. Leaf expansion was significantly reduced ($p<0.01$) by increasing concentration of salt in soil. A negative relationship was obtained between leaf area and salt concentration ($p<0.01$).

Effect of salinisation on dry weight

Dry weight significantly decreased ($p<0.01$) for leaves, stems, shoots (leaves + stems), tap roots, lateral roots and total roots of seedlings in response to increasing concentration of salt (Table 1). A negative relationship was obtained between dry weight of tissues (leaves, stems, shoots, tap

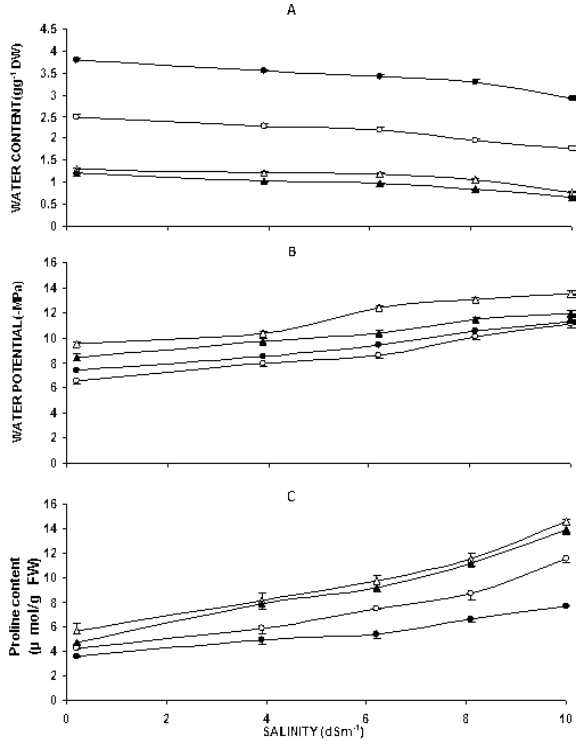

Figura 2. Efecto de la salinidad del suelo sobre **A:** contenido hídrico (gg^{-1} DW); **B:** agua potencial (-Mpa); **C:** contenido de prolina (μ mol/g FW) de hojas(○), tallo(Δ), raíz principal (▲) y raíces laterales (●) de plántulas de *Acacia senegal*. Las barras de error representan el SE.

Figure 2. Effect of soil salinity on **A:** water content (gg-1 DW); **B:** water potential (-Mpa); **C:** proline content (μ mol/g FW) of leaves (○), stem (Δ), tap root (▲) and lateral roots (●) of *Acacia senegal* seddling.

roots, lateral roots and total roots) and salt concentration ($p<0.01$).

Effect of salinisation on dry weight

Dry weight significantly decreased ($p<0.01$) for leaves, stems, shoots (leaves + stems), tap roots, lateral roots and total roots of seedlings in response to increasing concentration of salt (Table 1). A negative relationship was obtained between dry weight of tissues (leaves, stems, shoots, tap roots, lateral roots and total roots) and salt concentration ($p<0.01$). Percent relative weight of tissues of salinised plants compared to those of control plants were computed as: (salinised tissue dry weight / control dry weight) × 100. Dry weight values of tissues given in Table 1 were used for the calculation of percentage relative weight of tissues. Values of percent relative weight varied from 87.2 to 54% for leaves, from 78.1 to 50.2% for stems, 77.5 to 41.3% for tap roots and from 76 to 39.7% for lateral roots in response to increasing soil salinity from 3.9 to 10.0dSm^{-1}. As has been estimated using regression equations given in results, the salt concentration at which dry weight will be reduced to 50% of control plants (DW_{50}) were around 10.8, 9.3, 7.9 and 7.7 for leaves, stems ,tap roots and lateral root tissues, respectively. Root/shoot dry weight ratio was 1.10 under control conditions, while it was 1.05, 0.93, 0.89 and 0.87 for seedlings grown in soils at 3.9, 6.2, 8.1 and 10.0 dSm^{-1} salinities, respectively. Root/shoot dry weight ratios significantly decreased ($p<0.01$) as soil salinity increased. There was a negative relationship between root/shoot dry weight ratio and soil salinity ($r=-0.665$, $p<0.01$).

Effect of salinisation on water content of tissues

Water content in leaves, stems, taproots and lateral root tissues significantly decreased ($p<0.01$) with increasing concentration of salt in soil (Fig. 2A). There was maximum water content in lateral roots and minimum in tap roots. Tissues according to their water content can be arranged in following decreasing order: lateral roots >leaves > stems > tap roots. There was a negative relationship between water content in different tissues and salt concentration ($r=-0.744$, -0.770, -0.872 and -0. 793, $p < 0.01$, for leaves, stems, tap roots and lateral roots, respectively).

Effect of salinisation on water potential of tissues

Water potential significantly became more negative in leaves, stems, tap roots and lateral root tissues ($p<0.01$) as soil salinity increased (Fig. 2B). Tissues according to their water potential values (low to high negative) can be arranged in the following order: leaves > lateral roots > tap roots > stems. There was a negative relationship between water potential of tissues and salt concentration ($r=-0.961$, -0.906, -0.933 and -0.980, $p<0.01$, for for leaves, stems, tap roots and lateral roots, respectively).

Effect of salinisation on proline content of tissues

Proline content (μ mol/g FW material) significantly increased ($p<0.01$) in leaves, stems, tap roots and lateral root tissues with increase in soil salinity (Fig. 2C) .Tissues according to their pro-

Salinity (dSm^{-1})	Shoot height (cm)	Root length (cm)	Leaf area ($cm^2 plant^{-1}$)	Leaf weight ($mg\ plant^{-1}$)	Stem weight ($mg\ plant^{-1}$)	Shoot weight (leaf+stem) ($mg\ plant^{-1}$)	Tap root weight ($mg\ plant^{-1}$)	Lateral root weight ($mg\ plant^{-1}$)	Total root weight ($mg\ plant^{-1}$)
0.2	34.4±0.7	46.5±0.9	105.7±3.0	620.3±19.8	1880.6±19.1	2500.9±34.4	1674.5±42.8	1063.3±33.5	2737.8±57.9
3.9	29.3±0.7	38.5±0.8	85.2±2.8	540.8±17.5	1469.0±29.3	2009.8±30.8	1298.6±38.5	807.9±35.1	2106.5±50.6
6.2	25.6±0.8	30.5±0.7	66.3±3.0	455.7±11.9	1182.7±17.4	1638.4±25.3	930.3±33.6	592.5±27.3	1522.8±50.8
8.1	23.0±0.7	26.8±0.7	56.0±2.2	380.7±8.1	1031.7±18.2	1412.4±21.0	782.9±33.4	476.1±23.2	1259.0±42.7
10.0	19.3±0.4	24.5±0.9	46.8±2.5	334.7±8.9	944.6±14.4	1279.3±16.9	691.2±15.6	422.2±26.8	1113.4±34.9
α	34.97	46.77	107.17	638.57	1862.80	2501.40	1675.30	1062.30	2737.60
β	-1.52	-2.36	-6.18	-30.30	-98.78	-129.09	-105.60	-68.64	-174.25
r	-0.876	-0.924	-0.888	-0.885	-0.965	-0.941	-0.932	-0.896	-0.947
$LSD_{0.05}$	5.0	5.5	18.2	92.2	133.3	173.3	223.3	193.5	314.7

Relationship is significant at $p < 0.01$.

Tabla 1. Efecto de salinización del suelo en las características de hoja, tallo, brote y raíz de *Acacia senegal* indicado por la media ± DS y las constantes de la ecuación de regresión.

Table 1. Effect of salinisation of soil on leaf, stem, shoot and root characteristics of *Acacia senegal* as indicated by mean ± SEM and regression equation constants.

line content can be arranged in following decreasing order: stems > tap roots > leaves > lateral roots. There was a positive relationship between salt concentration and proline content of tissues(r=0.949, 0.953, 0.977 and 0.954, p<0.01, for leaves, stems, tap roots and lateral roots, respectively). A negative relationship was obtained between water potential and proline content (r=-0.954, -0.829, -0.919 and -0.963, p<0.01, for leaves, stems, tap roots and lateral roots, respectively). Similarly, a negative relationship was obtained between water content and proline content (r=-0.986, -0.956, -0.998 and -0.986, p<0.01, for leaves, stems, tap roots and lateral roots, respectively).

Effect of salinisation on mineral accumulation

Potassium and sodium content and K/Na ratio

Potassium content (as mg g^{-1} dry weight) significantly decreased (p<0.01) in leaves, stems, tap roots and lateral root tissues in response to increasing soil salinity (Table 2). There was a negative relationship between potassium content in tissues and increase in salt concentration in soil (p<0.01). Sodium content significantly increased (p<0.01) in leaves, stems, tap roots and lateral root tissues with increasing soil salinity. A positive relationship was obtained between Na content in tissues and increase in salt stress (p<0.01). K/Na ratio significantly decreased (p<0.01) in leaves, stems, tap roots and lateral roots in response to increase in soil salinity. A negative relationship was obtained between K /Na ratio in tissues and increase in salt stress (p<0.01).

Nitrogen, phosphorus, calcium and magnesium

Concentration of N, K and Ca was, in general, greater than that of P, Mg and Na in all tissues under control and salt stress conditions. Nitrogen, phosphorus, potassium and calcium content significantly decreased in leaves, stems, tap roots and lateral root tissues (p<0.01), as the salinity increased (Table 2). A negative relationship was obtained in N, P, K and Ca content of tissues and salt concentration (p<0.01). Magnesium content exhibited a significant increase (p<0.01) in leaves, stems, taproots and lateral root tissues in response to increase in salt stress. There was a significant positive relationship between Mg content in tissues and salt concentration in soil (p<0.01).

Micro–elements

There was a significant decrease in the concentration of Zn, Cu, Mn and Fe (p<0.01) in leaves, stems, tap roots and lateral root tissues in response to increase in salt stress (Table 2) . A negative relationship was obtained between soil salinity and Zn, Cu, Mn and Fe content in tissues (p<0.01).

Discussion

Earlier work (Ramoliya et al. 2004), indicated that seedling emergence for salt-tolerant legume tree *Acacia catechu* was reduced to 50% (SG_{50}) in soil with salinity of 6.0 dSm^{-1}, but for *Acacia senegal* SG_{50} was obtained at 5.9 dSm^{-1}. That would suggest that this plant species is relatively salt toler-

ant at seed germination. Under field conditions in coastal region of Saurashtra and in saline desert of Kutch, where this tree species grows, maximum soil salinity is found during the dry period and minimum during the rainy season (wet period) in the year. In general, salinity for the surface soil (0–15 cm depth) varies from 2.0 to 5.0 dSm^{-1}. Eventually, seeds of *A. senegal* can germinate and achieve establishment during the rainy season. However, salt concentration exceeding 10.0dSm^{-1} was detrimental to seed germination that can be attributed to decreasing osmotic potential of the soil solution. It was observed that seeds became non-viable within a few days in the soil with high concentration of salt. Although the effects of high salt content on metabolic processes are yet to be fully elucidated, it has been reported that salinity reduces protein hydration (Slater et al. 2003) and induces changes in the activities of many enzymes (Dubey & Rani 1990) in germinating seeds.

Reduction in water content and water potential of leaves, stems, tap roots and lateral roots of seedlings grown in saline soil might have resulted internal water deficit to plants, which in turn, reduced the growth of shoots and roots. It is found that plants subjected to water stress show a general reduction in size and dry matter production (Taiz & Zeiger 2006). Moreover, tap root elongation for seedlings grown in control and saline soils both was greater than that of shoot. Result suggests that this species has a tendency for rapid root extension. It is suggested that rapid root extension ensures existence of plants in dry habitats (Etherington 1987) and is considered an adaptation to survive in dry habitats. Root/shoot dry weight ratio of *A. senegal* was 1.1 under control conditions and was greater than that for aridity and salt tolerant seedlings of *A. catechu* (0.47) growing abundantly in saline desert of Kutch (Ramoliya et al. 2004).

In general, salinity can reduce plant growth or damage the plants through: (i) osmotic effect (causing water deficit), (ii) toxic effect of ions and (iii) imbalance of the uptake of essential nutrients. These modes of action may operate on the cellular as well as on higher organizational levels and influence all the aspects of plant metabolism (Kramer 1983, Garg & Gupta 1997). Results for reduction of shoot growth and leaf area development of *A. senegal* with increasing salt concentration are in conformity with the finding of Curtis & Lauchli (1986), who reported that growth in Kenaf (*Hibiscus cannabinus*) under moderate salt STRESS was affected primarily through a reduction in elongation of stem and leaf area development. Garg & Gupta (1997) reported that salinity causes reduction in leaf area as well as in rate of photosynthesis, which together result in reduced crop growth and yield. Also high concentration of salt tends to slow down or stop root elongation (Kramer 1983) and causes reduction in root production (Garg & Gupta 1997).

Results for dry weight and relative dry weight of tissues in response to increasing salinity suggest that there was maximum reduction in dry weight of tap roots and lateral roots while lowest in leaves. Consequently, leaves were more resistant than roots to increasing soil salinity. Tissues can be arranged in decreasing order of salt tolerance as: leaves>stems>tap roots=lateral roots. The greater reduction in root weight than in shoot weight resulted in decreased root/shoot dry weight ratio for seedlings with increase in salinity. In principle, salt tolerance can be achieved by salt exclusion or salt inclusion (Marschner 1995). The salt excluders exhibit water deficit which reduces the plant growth. Adaptation by exclusion requires mechanisms for avoidance of an internal water deficit. Adaptation by salt inclusion requires either high tissue tolerance to Na^+ and Cl^- or avoidance of high tissue concentration. The includers (halophytes) utilize inorganic salts (K^+, Na^+) for turgor maintenance or for the replacement of potassium ions in various metabolic functions by Na^+ (Marschner 1995). Consequently, growth of these plants does not decline under natural conditions and plants are salt tolerant. In the present study, seedlings of *A. senegal* survived up to the soil salinity of 10 dSm^{-1} and, therefore, this tree species is moderate salt tolerant. In addition, salinity caused reduction in growth of seedlings primarily through lowering the water status (or causing water deficit) in tissues. It is reported that salt excluders suffer from adverse effects on water balance and exhibit much reduced growth rates on saline substrates (Greenway & Munns 1980). As a result, this tree species can be grouped among salt excluders. Further, salt exclusion is a predominant salt avoidance mechanism in glycophytes (Greenway & Munns 1980). Considering selectivity of ions by root cells, it is still unclear which cell types control the selectivity of ions from the soil

Tissue	Salinity (dSm^{-1})	N ($mg\ g^{-1}$)	P ($mg\ g\text{-}^{1}$)	K ($mg\ g^{-1}$)	Na ($mg\ g^{-1}$)	Ca ($mg\ g^{-1}$)	Mg ($mg\ g^{-1}$)	K/Na ratio	Zn ($\mu g\ g^{-1}$)	Cu ($\mu g\ g^{-1}$)	Mn ($\mu g\ g^{-1}$)	Fe ($\mu g\ g^{-1}$)
Leaf	0.2	27±1.0	2.4±0.1	10.5±0.4	3.8±0.3	14.9±0.1	5.9±0.1	2.8±0.2	114±4.1	38±2.3	103±3.5	796±5.0
	3.9	25±1.1	2.0±0.1	9.3±0.4	4.8±0.3	11.5±0.4	6.0±0.1	2.0±0.1	43±4.6	34±2.2	88±4.3	515±5.1
	6.2	24±1.1	1.6±0.1	9.2±0.1	7.2±0.3	10.2±0.3	6.1±0.0	1.3±0.0	38±2.6	28±2.6	86±2.6	419±4.3
	8.1	23±0.8	1.5±0.0	7.0±0.5	9.0±0.2	8.3±0.5	6.2±0.1	0.8±0.0	28±4.7	26±2.0	82±1.5	405±4.7
	10.0	20±0.6	1.2±0.2	6.3±0.4	10.8±0.2	8±0.5	6.8±0.1	0.6±0.0	27±3.6	24±1.3	59±2.5	353±4.0
	α	27.51	2.43	10.96	2.92	14.71	5.75	2.84	98.69	38.58	105.45	747.95
	ß	-0.65	-0.12	-0.43	0.73	-0.72	0.07	-0.23	-8.57	-1.51	-3.84	-44.07
	r	-0.824	-0.913	-0.884	0.958	-0.958	0.733	-0.959	-0.883	-0.850	-0.777	-0.948
	$LSD_{0.05}$	2.9	0.4	0.4	0.9	1.3	0.4	0.5	11.8	6.3	9.0	13.7
Stem	0.2	22±1.1	2.0±0.1	6.6±0.2	2.1±0.1	10.9±0.3	3.1±0.2	3.2±0.2	138±3.2	35±2.0	50±1.5	780±3.5
	3.9	20±1.0	1.6±0.1	5.6±0.2	2.8±0.2	10.6±0.3	3.7±0.3	2.0±0.1	55±5.5	32±2.6	45±1.1	676±6.1
	6.2	18±1.5	1.2±0.1	4.8±0.2	4.5±0.3	9.4±0.4	4.0±0.3	1.1±0.1	46±3.6	28±2.5	38±1.7	556±3.6
	8.1	16±1.1	1.1±0.0	3.9±0.2	4.7±0.3	9.1±0.4	4.3±0.2	0.8±0.1	40±4.1	24±1.7	29±2.0	480±3.7
	10.0	15±0.6	0.8±0.2	3.8±0.3	5.2±0.2	7.1±0.5	5.6±0.5	0.7±0.1	35±4.3	23±0.7	24±2.6	361±5.5
	α	22.44	2.05	6.69	1.93	11.48	2.85	3.05	119.83	35.91	52.91	812.29
	β	-0.74	-0.12	-0.30	0.33	-0.36	0.22	-0.26	-10.04	-1.32	-2.76	-42.55
	r	-0.845	-0.919	-0.930	0.955	-0.829	0.795	-0.948	-0.884	-0.829	-0.939	-0.988
	$LSD_{0.05}$	3.3	0.4	0.8	0.4	1.3	1.0	0.4	12.4	6.0	5.6	13.6

Relationship is significant at $p < 0.01$

Tabla 1-Parte 1ª (Hoja & tallo). Efecto de la salinización del suelo sobre el contenido de nutrientes de los tejidos (hoja, tallo, raíz primaria y raíces laterales) de *Acacia Senegal* indicado indicado por la media ± DS y las constantes de la ecuación de regresión.

Table 2-Part 1 (Leaf & stem). Effect of salinisation of soil on nutrient content of tissues (leaf, stem, tap root and lateral root) of *Acacia Senegal* as indicated by mean ± SEM and regression equation constants.

Tissue	Salinity (dSm^{-1})	N ($mg\ g^{-1}$)	P ($mg\ g^{-1}$)	K ($mg\ g^{-1}$)	Na ($mg\ g^{-1}$)	Ca ($mg\ g^{-1}$)	Mg ($mg\ g^{-1}$)	K/Na ratio	Zn ($\mu g\ g^{-1}$)	Cu ($\mu g\ g^{-1}$)	Mn ($\mu g\ g^{-1}$)	Fe ($\mu g\ g^{-1}$)
Tap roots	0.2	18±1.0	1.6±0.1	5.0±0.1	2.0±0.2	8.8±0.3	2.8±0.2	2.6±0.3	131±4.1	32±1.5	68±2.5	3646±5.8
	3.9	16±0.6	1.2±0.1	4.5±0.2	2.3±0.1	6.8±0.5	3.3±0.3	2.0±0.2	59±4.9	22±1.1	55±1.5	3460±5.5
	6.2	15±0.7	1.0±0.1	3.0±0.2	3.7±0.2	4.9±0.4	4.1±0.2	0.8±0.1	36±3.0	20±1.7	41±3.6	3227±3.6
	8.1	14±0.5	0.7±0.1	2.8±0.3	4.4±0.3	4.8±0.4	4.3±0.3	0.7±0.1	34±3.7	17±2.0	29±1.1	3011±4.9
	10.0	12±0.7	0.6±0.1	2.5±0.2	5.4±0.1	3.6±0.5	5.1±0.4	0.5±0.0	31±4.3	15±0.5	25±1.7	2930±5.0
	α	18.29	1.61	5.15	1.51	8.77	2.61	2.63	115.79	30.81	70.14	3697.20
	β	-0.58	-0.10	-0.28	0.35	-0.52	0.23	-0.23	-10.14	-1.69	-4.67	-77.88
	r	-0.865	-0.893	-0.904	0.932	-0.928	0.850	-0.911	-0.904	-0.917	-0.971	-0.987
	$LSD_{0.05}$	2.3	0.4	0.7	0.6	1.3	0.9	0.6	12.0	4.4	6.7	14.8
Lateral roots	0.2	19±1.0	1.5±0.0	4.3±0.2	4.1±0.1	8.4±0.4	4.5±0.2	1.1±0.1	148±4.0	76±1.5	175±2.6	5225±4.5
	3.9	15±1.0	1.2±0.1	4.2±0.1	5.2±0.2	7.2±0.5	5.2±0.3	0.8±0.1	77±3.5	67±1.0	164±2.0	5016±5.7
	6.2	14±1.1	0.8±0.1	4.0±0.1	6.1±0.3	6.5±0.3	5.9±0.3	0.7±0.0	47±4.9	60±2.6	159±3.0	4743±5.1
	8.1	13±1.0	0.6±0.0	3.5±0.2	7.8±0.5	6.4±0.6	6.0±0.2	0.5±0.0	44±4.6	56±1.1	133±3.2	4572±5.5
	10.0	12±0.5	0.4±0.1	3.1±0.1	7.9±0.5	3.0±0.5	6.6±0.2	0.4±0.0	41±4.1	50±2.5	124±0.5	4543±4.5
	α	18.54	1.56	4.51	3.82	8.94	4.44	1.07	134.02	76.78	181.43	5251.30
	β	-0.69	-0.11	-0.12	0.42	-0.46	0.21	-0.06	-11.01	-2.63	-5.35	-75.96
	r	-0.848	-0.927	-0.808	0.917	-0.821	0.868	-0.934	-0.918	-0.957	-0.930	-0.982
	$LSD_{0.05}$	2.9	0.3	0.5	1.1	1.5	0.8	0.2	12.5	5.6	7.3	15.0

Relationship is significant at $p < 0.01$

Tabla 1-Parte 2ª (Raíces). Efecto de la salinización del suelo sobre el contenido de nutrientes de los tejidos (hoja, tallo, raíz primaria y raíces laterales) de *Acacia Senegal* indicado indicado por la media ± DS y las constantes de la ecuación de regresión.

Table 2-Part 2 (Roots). Effect of salinisation of soil on nutrient content of tissues (leaf, stem, tap root and lateral root) of *Acacia Senegal* as indicated by mean ± SEM and regression equation constants.

solution.

In some plant species, salt tolerance associates with accumulation of organic solutes in cytoplasm to balance the osmotic pressure of ions in the vacuoles. The compounds that accumulate most commonly are proline and glycine betaine, although other molecules can accumulate to high concentration in certain species (Hasegawa et al. 2000). Proline accumulates in the cytoplasm without having any detrimental effects on cytosolic enzymes activities (Stewart & Lee 1974). In addition, the primary role of proline may not be solely as an osmolyte, but it also helps the cells to overcome oxidative stress in salt stressed plants (Rajendrakumar et al. 1994).

The cation K^+ is essential for cell expansion, osmoregulation and cellular and whole–plant homeostasis (Schachtman et al. 1997). High stomatal K^+ requirement is reported for photosynthesis (Chow et al. 1990). The role of K^+ in response to salt stress is also well documented, where Na^+ depresses K^+ uptake (Fox & Guerinot 1998). In the present study, significant decrease of K^+ content in all the tissues of seedlings with increasing soil salinity suggests that Na^+ inhibited K^+ uptake.

The exchange of K^+ for Na^+ by the cells in the stele of the roots or in the vascular bundles in stems is considered as one type of control to transport of salts to leaves or growing tissues. Moreover, the significant increase of Na to leaves and stem tissues of *A. senegal* suggests that this mechanism to block Na+ transfer to growing tissues was not effective at high salt concentration. The decrease in K^+/Na^+ ratio in all the tissues with increase in salinity can be accounted for relatively low accumulation of K^+ than that of Na^+. As a consequence there were no effective mechanisms to control net uptake of Na+ on root plasma membrane and subsequently its transport to shoot tissue. The pattern of accumulation of K^+ and Na^+ in *A. senegal* conforms to group C and/or group D plants in Marschner's (1995) classification of the ability of plants to substitute Na^+ with K^+. In this classification Marschner divided plants into four groups, A, B, C and D depending upon whether K^+ is mostly exchangeable with Na^+. Sodium has a positive effect on growth in A and B plants (mostly salt tolerant plants). Group C plants contain very little K^+ that can be substituted with Na without a negative effect on growth, and group D plants exhibit no K^+/Na^+ substitution (salt-sensitive plants).

It is reported that uptake mechanisms of both K^+ and Na^+ are similar (Watad et al. 1991, Schroeder et al. 1994). Plants utilize two systems for K^+ acquisition, low- and high-affinity uptake mechanisms. Na^+ can not move through the plasma membrane lipid bilayer, but the ion is transported through both low- and high–affinity transport systems, which are necessary for K^+ acquisition. As a consequence, Na^+ could enter the cell through high affinity K^+ carriers or through the low affinity channels called non selective cation channels that are strongly influenced by Ca_2^+. These cation channels could allow entry of large amount of Na^+ from a highly saline soil if not adequately regulated (Amtmann & Sanders 1999). Low affinity K^+ uptake is not inhibited by Na^+ but the high affinity process is restricted (Watad et al. 1991, Schroeder et al. 1994). Similarly Na^+ toxicity in plants is correlated with two proposed Na^+ uptake pathways (Maathuis & Sanders 1994, Niu et al. 1995). The K^+ and Na^+ profiles of *A. senegal* suggest that similar mechanism might operate in this species. It is evidenced that Ca^{2+} causes closure of nonselective cation channels and restricts Na^+ uptake (Rus et al. 2001). As a result, calcium fertilizers may mitigate Na^+ toxicity to this plant.

In general, salinity reduces N accumulation in plants (Feigin 1985). This is due to the fact that an increase in chloride uptake and accumulation is mostly accompanied by a decrease in shoot nitrate concentration (Torres & Bingham 1973, Garg & Gupta 1997). The Interaction between salinity and P is very complex and there is no clear cut mechanistic explanation for decreased, increased or unchanged P uptake in response to salinisation in different species (Champagnol 1979, Grattan & Grieve 1992). However it is known that P concentration is related to the rate of photosynthesis, since it decreases the conversion of fixed carbon in to starch (Overlach et al. 1993) and therefore decrease of P in leaves will reduce shoot growth.

Calcium is important during salt stress, e.g., in preserving membrane integrity (Rengel 1992), signaling in osmoregulation (Mansfield et al. 1990) and influencing K^+/Na^+ selectivity (Cramer et al. 1987). In the present study, there was a significant decrease of Ca^{2+} content in all the tissues with salinisation of soil. As a result, Na^+ induced Ca^{2+} deficiency in tissues. It is reported that up-

take of Ca^{2+} from the soil solution may decrease because of ion interactions, precipitation and increase in ionic strength that reduce the activity of Ca^{2+} (Janzen & Chang 1987, Garg & Gupta 1997). Besides the role of Mg^{2+} in chlorophyll structure and as an enzyme cofactor, another important role of Mg^{2+} in plants is in the export of photosynthates (Marschner & Cakmak 1989). In the present study increase of Mg^{2+} in tissues may be of importance for plant growth and survival in saline soils.

It is difficult to suggest mechanistic explanations of salinity influence on micro–element concentration due to relatively smaller differences between control and salinised tissues (Tozlu et al. 2000). In the present study, it appears that salinity reduced Zn, Cu, Mn and Fe accumulation, at the whole plant level. Besides, cofactors for enzymes, Fe and Cu are essential for biological redox system, Mn for photolysis of water in photosynthesis and Zn for DNA replication, regulation of gene expression and integrity of biomembranes (Marschner 1995). In addition, high concentration of iron is required for structural and functional integrity of the thylakoid membranes and synthesis of ferredoxin and chlorophyll (Marschner 1995). Pushnik and Miller (1989) reported that iron is involved in photosystem I (PSI) development and assembling the subunits in the thylakoid membranes. The simultaneous decrease of Zn, Cu, Mn and Fe in leaves of *A. senegal* might limit the growth of plants. Salinity generates an increase in reactive oxygen species (ROS) which have deleterious effects on cell metabolism (Borsani et al. 2001). Super oxide dismutases (SODs) detoxify ROS and may contain Cu, Zn, Mn and Fe as metal components (Slater et al. 2003). Decrease in Cu, Zn, Mn and Fe content at the whole plant level might affect the survival of *A. senegal* in saline soil where salinity exceeded 10.0dSm^{-1}.

References

Amtmann A. & Sanders D. 1999. Mechanisms of Na^+ uptake by plant cells. Advances in Botanical Research 29:76-112.

Bates LS, Waldren RP & Teare FD. 1973. Rapid determination of free proline from water stress studies, Plant and Soil 39:205-207.

Bernstein L. 1962. Salt affected soils and plants. Proceedings of the Paris Symposium, UNESCO (May 1960). Arid Zone Research 18: 139-174.

Borsani O, Valpuesta V & Botella MA. 2001. Evidence for a role of salicylic acid in the oxidative damage generated by NaCl and osmotic stress in Arabidopsis seedlings. Plant Physiology 126:1024-1030.

Champagnol F.1979. Relationship between phosphate nutrition of plants and salt toxicity. Phosphorus Agriculture 76:35-43.

Chow WS, Ball MC & Anderson JM .1990. Growth and photosynthetic responses of spinach to salinity, implication of K nutrition for salt tolerance. Australian Journal of Plant Physiology 17: 563-578.

Cramer GR, Epstein E & Lauchli A. 1989. Na-Ca interactions in barley seedlings: relationship to ion transport to growth. Plant cell and Environment 12: 551-558.

Cramer GR, Lynch J, Lauchli A, Polito US. 1987. Influx of Na^+, K^+ and Ca into roots of salt-stressed cotton seedlings. Effects of supplemental Ca. Plant Physiology 83: 510-516.

Curtis PS & Lauchli A .1986. The role of leaf area development and photosynthetic capacity in determining growth of Kenaf under moderate salt stress. Australian Journal of Plant Physiology 13: 553-565.

Dubey RS & Rani M. 1990. Influence of NaCl salinity on the behavior of protease, aminopeptidase and carboxyl-peptidase in rice seedlings in relation to salt tolerance. Australian Journal of Plant Physiology 17: 215-24.

Etherington JR. 1987. Penetration of dry soil by roots of Dactylis glomerata L. clones derived from well drained and poorly drained soils. Functional Ecology 1:19-23.

Feigin A.1985. Fertilization management of crops irrigated with saline water. Plant Soil 89: 285-299

Fox TC and Guerinot ML. 1998. Molecular biology of cation transport in plants. Annual Review of Plant Physiology and Plant Molecular Biology 49: 669-696.

Garg BK & Gupta IC. 1997. Saline Wastelands Environment and Plant Growth. Jodhpur: Scientific Publishers, pp.283.

Grattan SR & Grieve CM. 1992. Mineral element acquisition and growth response of plants grown in saline environments. Agriculture Ecosystem and Environment 38: 275-300.

Greenway H & Munns R. 1980. Mechanisms of salt tolerance in nonhalophytes. Annual Review of Plant Physiology 31: 149-190.

Hasegawa PM, Bressan RA, Zhu JK & Bohnert HJ. 2000. Plant cellular and molecular responses to high salinity. Annual Review of Plant Physiology and Plant Molecular Biology 51: 463-499.

Janzen HH & Chang C.1987. Cation nutrition of barley as influenced by soil solution composition in a saline soil. Canadian Journal of Soil Science 67: 619-629.

Kramer PJ.1983. Water Relations of Plants. New York: Academic Press pp .489.

Maas EV & Grieve CM.1987. Sodium induced calcium deficiency in salt–stressed corn. Plant Cell and Environment 10:559-564.

Maas EV & Hoffman GJ.1977. Crop salt tolerance-current assessment. Journal of Irrigation Drainage Division ASCE 103: 115-134.

Maathuis FJM & Sanders D.1994. Mechanism of high-affinity potassium uptake in roots of Arabidopsis

thaliana. Proceedings of the National Academy of Sciences USA 91: 9272-9276.

Mansfield TA, Hetherington AM & Atkinson CJ. 1990. Some aspects of stomatal physiology. Annual Review of Plant Physiology and Plant Molecular Biology 41: 55-75.

Marschner H .1995. Mineral Nutrition of Higher Plants London: Academic Press, pp 889.

Marschner H. & Cakmak I .1989.High light intensity enchances chlorosis and necrosis in leaves of zinc, potassium and magnesium deficient bean Phaseolus vulgaris plants. Journal of Plant Physiology 134: 308-315.

Munns R.2002. Comparative physiology of salt and water stress. Plant Cell and Environment 25: 239-250.

Munns R.1993. Physiological processes limiting plant growth in saline soils: some dogmas and hypotheses. Plant cell and Environment 16: 15-24.

Niu X, Bressan RA, Hasegawa PM & Pardo JM. 1995. Ion homeostasis in NaCl stress environments. Plant Physiology 109: 735-742.

Overlach S, Diekmann W & Raschke K. 1993. Phosphate translocator of isolated guard-cell chloroplasts from Pisum sativum L. transports glucose-6-phosphate. Plant physiology 101: 1201-1207.

Patel AD & Pandey AN (in press). 2007. Effect of salinisation of soil on growth, water status and nutrient accumulation in seedlings of Jatropha curcas(Euphorbiaceae). Basic Applied and Dry Land Research.

Piper CS. 1944. Soil and Plant Analysis. Interscience, New York, 368pp.

Pushnik JC & Miller GW. 1989. Iron regulation of chloroplast photosynthetic function: mediation of PSI development. Journal of Plant Nutrition 12:407-421.

Rajendrakumar CS, Reddy BV & Reddy AR. 1994. Proline protein interaction: Protection of structural and function integrity M4 lactatdyhydrogenase. Biochemistry Biophysics Research Communication. 201: 957- 963.

Ramoliya PJ, Patel HM & Pandey AN. 2006. Effect of salinization of soil on growth and nutrient accumulation in seedlings of Prosopis cineraria. Journal of Plant Nutrition 29: 283-303.

Ramoliya PJ, Patel HM & Pandey AN. 2004. Effect of salinization of soil on growth and macro-and micronutrient accumulation in seedlings of Acacia catechu (Mimosaceae). Annals of Applied Biology 144: 321-332.

Rengel Z.1992. The role of calcium in salt toxicity. Plant Cell and Environment 15: 625-632.

Rus A, Yokoi S, Sharkhuu A, Reddy M, Lee BH, Matsumoto TK, Koiwa H, Zhu JK, Bressan RA & Hasegawa PM. 2001. AtHKT1 is a salt tolerance determinant that controls Na^+ entry into plant roots. Proceedings of Natural Academy of Sciences USA 98: 14150-14155.

Schachtman DP, Kumar R, Schroeder JI & Marsh EL.1997. Molecular and functional characterization of a novel low-affinity cation tranporter (LCTI) in higher plants. Proceedings of the Natural Academy of Sciences USA 94: 11079-11084.

Schroeder JI, Ward JM & Gassmann W. 1994. Perspectives on the physiology and structure of inward-rectifying K channels in higher plants, biophysical implications for K uptake. Annual Review of Biophysics and Biomolecular Structure 23:441-471.

Slater A, Scott N & Fowler M. 2003. Plant Biotechnology. The genetic manipulation of Plants. New York: Oxford University Press Inc.

Stewart GR & Lee JA.1974. The role of proline accumulation in halophytes. Planta 120: 279-289.

Taiz L & Zeiger E. 2006. Plant physiology (Fourth Edition) Sunderland, USA: Sinauer Associates, Inc., Publishers, pp. 764.

Torres BC & Bingham FT. 1973. Salt tolerance of Mexican wheat. I. Effect of No_3 and NaCl on mineral nutrition, growth and grain production of four wheats. Soil Science society of America Proceedings 37: 711-715

Tozlu I, Moore GA & Guy CL. 2000. Effect of increasing NaCl concentration on stem elongation, dry mass production, and macro-and micro-nutrient accumulation in Poncirus trifoliata. Australian Journal of Plant Physiology 27: 35-42.

Watad AA, Reuveni M, Bressan RA & Hasegawa PM.1991. Enhanced net K uptake capacity of Nacl-adapted cells. Plant Physiology 95: 1265-1269.