ANNALS OF PLANT SCIENCES
ISSN: 2287-688X
OPEN ACCESS
www.annalsofplantsciences.com

ORIGINAL RESEARCH ARTICLE

Variation in mineral contents of *Ceriops decandra* (Griff.) Ding Hou under NaCl stress-A true mangrove species

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Received: November 16, 2016; **Revised:** November 25 2016; **Accepted:** December 15, 2016

Abstract: 30 days old healthy seedlings of *Ceriops decandra* were selected for NaC1 treatment. The experimental plants were treated with NaCl solution (Control, 200, 400, 600, 800, 1000 mM) by soil drenching method to analyze the ions such as Sodium, Calcium, Chloride, Nitrogen, Phosphorous and Manganese. The results showed that above 800 mM the seedlings did not survived. Sampling for analysis of ions was taken on the 60th, 90th and 120th days after planting (DAP). Nitrogen and Calcium content were increased in 400 mM NaCl leaf tissues and were slightly increased in stem and roots of 400 mM. Chloride and Sodium was more accumulated by 800 mM NaCl treated leaves and then stem and root. Magnesium and Phosphorous content were decreased above the optimum concentration of NaCl.

Key words: *Ceriops*; ions; optimum; Salinity; Sodium.

Introduction

Soil salinity not only results in reduced growth of plants due to osmotic or ionic effects by Munns and Tester (2008) but also bring about several changes in physiological responses (Flowers *et al.*, 1977; Glenn *et al.*, 1999) and metabolic shift during the course of development. Salt induces osmotic stress by limiting absorption of water from soil, and ionic stress resulting from high concentrations of potentially toxic salt ions within plant cells. Plants have evolved a variety of protective mechanisms to allow with these unfavorable environmental conditions for survival and growth including the accumulation of ions and osmolytes such as Proline. The accumulation of these compounds prevents water loss and ion toxicity. The alleviation of oxidative damage and increased resistance to salinity and other environmental stresses are often correlated with an efficient Antioxidative system (Jaleel *et al.*, 2007 and Manivannan *et al.*, 2007). Salinization of agricultural soils is a worldwide concern, especially in irrigated lards. Saline soil is characterized by the presence of toxic levels of sodium and its chlorides and sulphates. Halophytes, plants that survive to reproduce in environments where the salt concentration is around 200 mM NaCl or more, constitute about 1% of the world flora by Flowers and Colmer (2008). Mangrove vegetation is generally dominated by halophytic woody plant species, and mangrove species are the most tolerant to both high and fluctuating water salinity, ranging from low in estuarine habitats to hyper saline conditions in sites regularly fed by seawater (Hajer *et al.*, 2006). As the mangrove, environmental conditions affect the survival and the productivity of the colonizing plants species, plant structures and physiological features explain their ecological success under harsh conditions (Clough *et al.*, 1982). Accumulation of inorganic ions in vacuoles is common pattern observed in mangrove plants under saline conditions (Ball MC and Farquhar GD 1984; Naidoo G 1987) which serves not only to increase cellular osmolarity to counter osmotic stress but also to avoid increases in ionic strength of the cytoplasm (Dasgupta *et al.*, 2012; Mimura *et al.*, 2003). However, previous studies on this topic mostly focused on a certain growth stage of mangroves, how salinity influences mangroves in a dynamic developmental process is not well known. The main objective of the present study is to analyze the ion contents of *Ceriops decandra* under NaCl salinity.

Materials and Methods

Collection of propagules: Propagules of *Ceriops decandra* belongs to the family Rhizophoraceae were collected from Pichavaram mangrove forest situated at 11° 27' N Latitude and 79° 47' E Longitude, in the East coastal region of Tamilnadu, India.

Plant materials and culture conditions: The propagules of *C. decandra* was planted and grown individually in polythene bags (7"× 5") filled with homogenous mixture of garden soil containing red soil, sand and farm yard manure (1:2:1). The propagules were irrigated with tap water and maintained in the Botanical Garden, Annamalai University.

Salt treatment and experimental design: 30 days old healthy seedlings were selected for NaCl treatment. The preliminary experiments were carried out in *C. decandra* at different concentrations of NaCl (200, 400, 600, 800, 1000 mM) by soil drenching method. Above 800 mM the seedlings did not survived. The experimental plants treated with NaCl by soil drenching method in alternate days up to 800mM were alone maintained in the experimental plot. The experimental yard was roofed with transparent polythene sheet at a height of 3m from the ground in order to protect the plants from rain. Sampling for ion determination studies was taken on the 60th, 90th and 120th days after planting.

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Determination of ion constituents

Samples were collected randomly on 60th, 90th and 120th day after planting. The seedlings were separated into leaves, stem and root and used for analysis of ions such as Sodium, Calcium, Chloride, Nitrogen, Phosphorous and Manganese. The sodium content was determined by the method of Willams and Twine (1960). The dried and ground tissues of 0.5g were digested in 100 ml Kjeldahl flasks using 10 ml of concentrated nitric acid, 0.5 ml of 60 per cent perchloric acid and 0.5 ml of concentrated sulfuric acid. Digestion was continued until the nitric acid and perchloric acid were driven off. The inorganic residue was cooled and diluted with 15ml of distilled water and filtered through Whatmann No. 42 filter paper. The filtrate was made up to 50ml with distilled water. The filtrates were used for sodium estimation and a Flame Photometer (Systronics, India) were used for the purpose and standards were prepared with sodium chloride. The calcium content was estimated by the method of Yoshida *et al.,* 1972. Two ml of the filtrate was mixed with two ml of 5% lanthanum oxide solution and diluted with ten ml of 1N hydrochloric acid. The solution was fed into an Atomic Absorption Spectrophotometer (Perkin Elmer-2280) at 211.9nm. Standard curve was prepared by using calcium chloride. Phosphorus Phosphorous was determined spectro-photometrically following Jackson 1962. Manganese was determined by the method of Celik and Katkat (2009). Nitrogen was determined by Kjeldahl digestion method according to Bremner 1965. Chloride was analyzed by precipitation as AgCl and titration according to Johnson and Ulrich (1959).

Statistical analyses

The pot experiment was set up in randomized complete block design replicated five times. ANOVA (SAS version 21.0) was employed for statistical analysis of data.

Results

The level of various ion contents was analyzed to get an insight into the effects of salinity on ion uptake and their accumulation in leaf, stem, and root of *Ceriops decandra.*

Effect of NaCl on Sodium content (mg/g dr. wt.) in the leaf, stem and root of *Ceriops decandra* at 60th, 90th and 120th days after planting

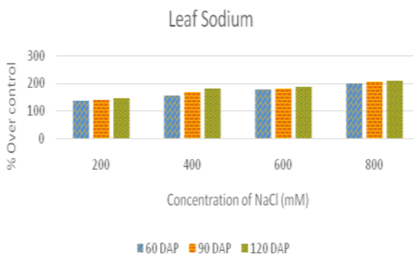

Figure 1

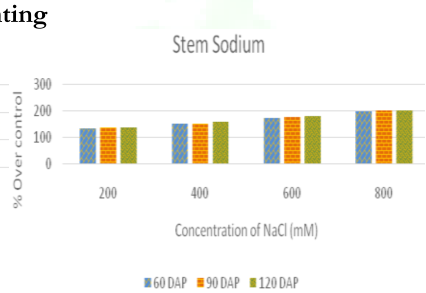

Figure 2

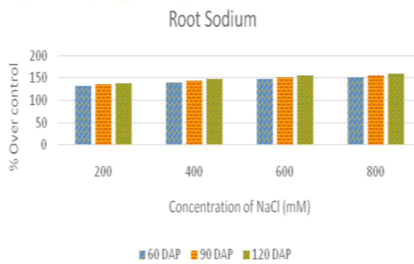

Figure 3

Effect of NaCl on Chloride contents (mg/g dr. wt.) in the leaf, stem and root of *Ceriops decandra* at 60th, 90th and 120th days after planting

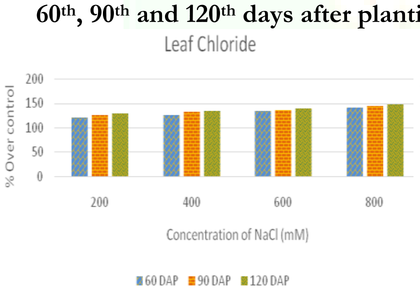

Figure 4

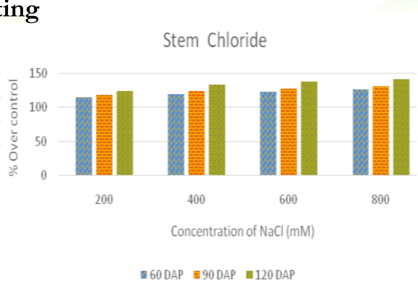

Figure 5

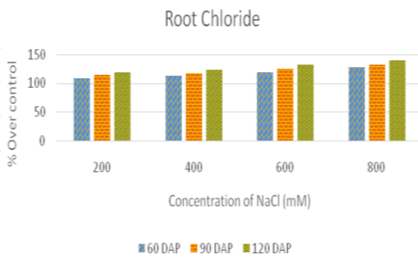

Figure 6

Effect of NaCl on Calcium contents (mg/g dr. wt.) in the leaf, stem and root of *Ceriops decandra* at 60th, 90th and 120th days after planting

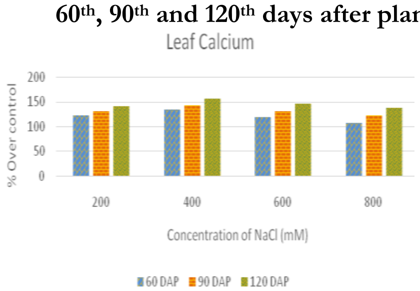

Figure 7

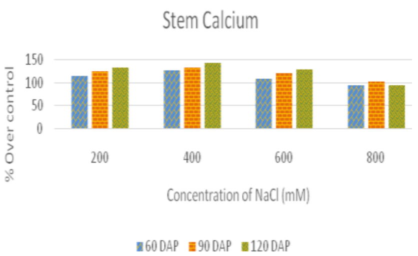

Figure 8

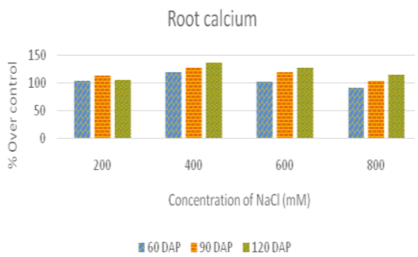

Figure 9

Effect of NaCl on Nitrogen contents (mg/g dr. wt.) in the leaf, stem and root of *Ceriops decandra* at 60th, 90th and 120th days after planting

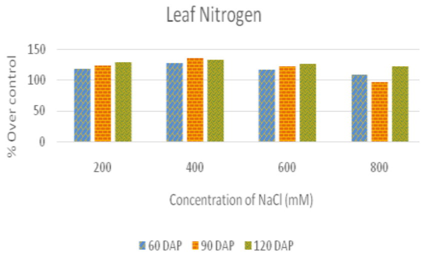

Figure 10

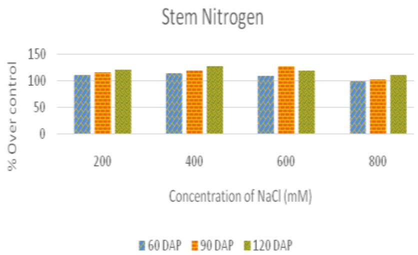

Figure 11

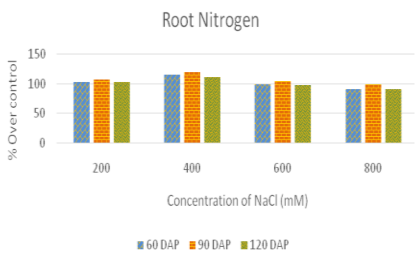

Figure 12

Effect of NaCl on Phosphorous contents (mg/g dr. wt.) in the leaf, stem and root of *Ceriops decandra* at 60th, 90th and 120th days after planting

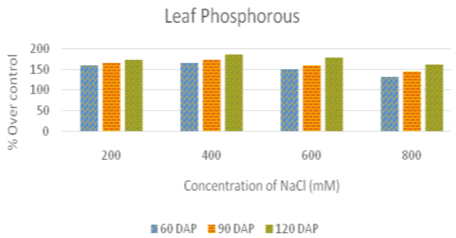

Figure 13

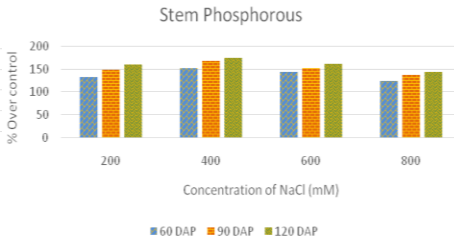

Figure 14

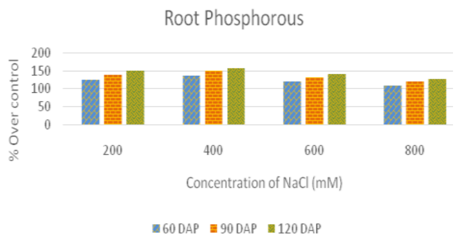

Figure 15

Effect of NaCl on Manganese contents (mg/g dr. wt.) in the leaf, stem and root of *Ceriops decandra* at 60th, 90th and 120th days after planting

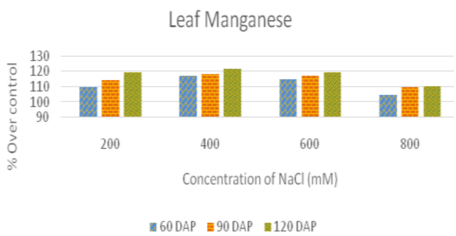

Figure 16

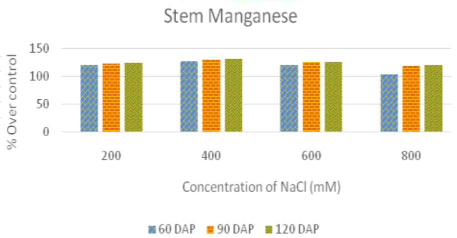

Figure 17

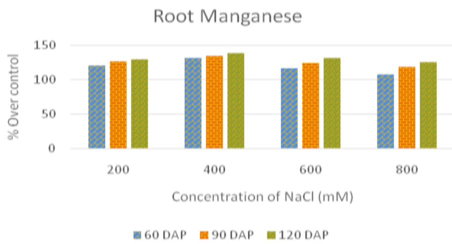

Figure 18

Sodium

Sodium content were observed to accumulation in response to NaCl salinity in leaf of *Ceriops decandra*. In leaves the accumulation of sodium content was found to higher than control on 60, 90 and 120 DAP in 800mM NaCl concentration. It was higher accumulation of sodium when compared to control and other treatments. The stem tissues showed higher accumulation of sodium content was found to be maximum than control on 120th DAP in 800mM NaCl concentration. The root tissues showed more accumulation of sodium ions when compared to control. The sodium content was observed as increased in 200, 400, 600 and 800 mM NaCl treated plants on 120th DAP. Among all sampling days and treatments, the 800 mM NaCl on 120th DAP showed more accumulation of sodium content

Chloride

In leaf of *C. decandra,* the chloride ion content was increased than control on 60th, 90th and 120th DAP in 800mM of NaCl stress respectively. The other NaCl treated plants were showed increased level accumulation of chloride in all sampling days but 800mM concentration showed highest accumulation of chloride content. In stem tissues the chloride ion accumulation was significantly increased then control on on 60th, 90th and 120th DAP in 800mM of NaCl treated plants. It was more when compared to control and other treatments. The root of *Ceriops decandra,* an uptake of chloride ions was observed under NaCl stress. The accumulation of chloride ions was found higher on 60th, 90th and 120th DAP of 800mM NaCl stress. In the leaf of C. decandra, chloride accumulation was observed to be higher than in their stem and root.

Calcium

In the leaves of *C. decandra*, a increase in calcium ion content was observed which was increased over to control under 400mM NaCl stress on 60th, 90th and 120th DAP in 800mM NaCl stress. When salt concentration was increased the accumulation of calcium ion was decreased when compared to other treatments. In the stems of *C. decandra*, calcium ion content increased in 400mM of NaCl stress on 60th, 90th and 120th DAP. In the roots of *C. decandra* calcium ions accumulation decreased when salt concentration was increased. In roots calcium ion accumulation was increased in 200mM and 400mM NaCl stress. But decreased in 600mM and 800mM of NaCl treated plants.

Nitrogen

The accumulation of nitrogen content was observed in leaf, stem and root of *Ceriops decandra.* In leaf the nitrogen accumulation was higher in 400mM of NaCl stress on 60th, 90th and 120th DAP. In 600mM and 800mM of NaCl treated plants showed increased percentage over the control and decreased when compared to other treatments. The stem tissues showed increased accumulation of nitrogen in 400mM NaCl concentration treated plants. The decreased accumulation was showed in 600mM and 800mM NaCl treated plants. The root of *C. decandra* showed more accumulation of nitrogen in 400mM NaCl concentration treated plants on 60th, 90th and 120th DAP. In 600mM and 800mM of NaCl treated plants showed increased percentage over the control and decreased when compared to other treatments.

Phosphorous

In leaf of Ceriops decandra, phosphorous ion accumulation exhibited a contrasting trend under NaCl stress. In increasing NaCl concentrations, potassium ion accumulation was observed to increase in the leaf of *Ceriops decandra* upto 400mM of NaCl stress respectively on 60th, 90th, and 120th DAP. In stem tissues of *Ceriops decandra* the accumulation of phosphorous ions was increased in 400mM of NaCl stress respectively on 60th, 90th, and 120th DAP. The root tissues of *Ceriops decandra* showed more accumulation of phosphorous was maximum in 400mM of NaCl stress respectively on 60th, 90th, and 120th DAP. When salinity increased above optimaum level the phosphorous content was decreased.

Manganese

The accumulation of manganese content was observed in leaf, stem and root of *Ceriops decandra.* The leaf tissues showed increased accumulation of manganese when plants treated with 400mM of NaCl concentration on 60th, 90th, and 120th DAP. In 600mM and 800mM of NaCl treated plants showed increased percentage over the control and decreased when compared to other treatments. The manganese content was more accumulated by stem tissues when treated with 400mM of NaCl concentration on 60th, 90th, and 120th DAP. In root of *Ceriops decandra,* when plants treated with 400mM NaCl concentration showed increased accumulation of manganese. The increased salinity was decreased the accumulation of manganese in root.

Discussion

The NaCl treatment increased the sodium content more in the leaf tissues than in the stem and root tissues with increasing of NaCl salinity up to extreme levels in *Ceriops decandra* at 60th, 90th and 120th day the sampling days. Increased availability of sodium in the soil influenced a proportional increase in the uptake of sodium by the seedlings. Many salt tolerant organisms accumulate higher intracellular ion concentration than their non-tolerant counter parts. Different halophytes have been reported to differ in the rate of accumulation of sodium in their cells and differences are mainly due to difference in the mode of salt regulation. Some vascular halophytes accumulate high levels of sodium and other salts in their above ground tissue while others did not (Gorham *et al.*, 1987). Increasing levels of NaCl induced a progressive absorption of Na and Cl in both shoot and root agreeing with the result of (Chavan and Karadge 1986; Turan *et al.*, 2007). Excessive Na concentration in the plant tissue hinders nutrient balance, osmotic regulation and causes specific ion toxicity (Katerji *et al.*, 2004; Arzani 2008). Accumulation of Cl in the root tissue is disruptive to membrane uptake mechanisms, and these results in increased translocation of Cl to the shoots (Yousif *et al.*, 1972). The accumulation of high sodium content in halophytes in the present study are in agreement with those of several other accumulating type of halophyte *Suaeda fruticosa* (Khan *et al.*, 2000). The concentration of Na^+ in the leaves of salinized plants was approximately 40-fold higher than that measured in non-salinized controls (Maggio *et al.*, 2003). While sodium is not considered to be a plant nutrient, it is essential for halophytes to accumulate salts to maintain turgor pressure and growth (Borox 2002), for survival in high salt (Wong *et al.*, 2006). Salinity influences total nitrogen (N) uptake (Evelin *et al.*, 2012) and as well as interferes at different stages of its metabolism (Gomes *et al.,* 2011). In addition, previous studies have reported that improved N nutrition may help to reduce the toxic effects of Na ions by reducing its uptake and this may indirectly help in maintaining the chlorophyll content of the plant (Evelin *et al.*, 2009). Increase in calcium content can be attributed to the overall performance of metabolic activity of plants treated with the salts up to the optimum concentrations. Calcium also serves to protect membrane damage and it plays a key role in the selective transport of potassium in the presence of excess of sodium, and thereby making a plant more salt tolerant (Epstein 1980). Increasing external calcium salinity decreased the Ca^{2+} contents in *Aegiceras corniculatum* (Shindle and Bhosale 1985); *Rhizophora mucronata* and *Avicennia officinalis* (Bhosale and Malik 1991) and *Allenrolfea occidentalis* (Bilquees *et al.*, 2000). In *Suaeda nudiflora*, salinity caused no change in Ca^{2+} content (Joshi and Iyengar 1987). In this present study, it appears that salinity enhanced manganese accumulation. This same result proposed by Marschner 1995. Increases level of Manganese contents at the whole plant level might be the requirement of this plant for survival and growth in response to salinity (Salter, Scott and Flower, 2003).

Conclusion

In conclusion, the observation on NaCl treated *Ceriops decandra* plant suggested that growth and minerals constituents increased up to optimal level of salinity and at higher salinity declined gradually.

Acknowledgement

The Authors are grateful to acknowledge the Professor and Head, Department of Botany, Annamalai University and Authorities of Annamalai University to provide necessary facilities during the study period.

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Cite this article as:
Arunprasath A. and Gomathinayagam M. Variation in mineral contents of Ceriops decandra Griff Ding (Hou) under NaCl stress. *Annals of Plant Sciences* 5.12 (2016): 1458-1462.
DOI: http://dx.doi.org/10.21746/aps.2016.12.001

Source of support: Nil.
Conflict of interest: None Declared