INVOLVEMENT OF LEAF PROLINE CONTENT AND MYCORRHIZAL FUNGAL SYMBIOSIS FOR THE SALINITY TOLERANCE OF MANGROVE PLANTS IN KOGGALA LAGOON, SRI LANKA.

IC Senanayake, PN Yapa* and RBN Dissanayake

Department of Biological Sciences, Faculty of Applied Sciences, Rajarata University of Sri Lanka, Mihintale.

* Pnyapa40@yahoo.co.ukU

INTRODUCTION

Mangroves are most specific ecosystem which is adapted to saline environment. Mangrove plants change their metabolism and form various adaptations to thrive in their environment when they are exposed to stress conditions. Salt stress changes the morphological, physiological and biochemical responses of mangroves. Further salt stress causes osmotic and ionic stress in plants. Production and accumulation of free amino acid, especially proline by plant tissue is an adaptive response to salt stress, proposed to act as a compatible solute that adjusts the osmotic potential in the cytoplasm and also plants form symbiotic association with arbuscular mycorrhizal fungi (AMF) as a result of osmotic stress (Sengupta and Chaudhuri 2002). Arbuscular mycorrhizal association enhances the water uptake of the plant. The present study was made to investigate the relationship among plant free proline content, soil characters, arbuscular mycorrhizal colonization and the involvement of these factors for salt stress tolerance of mangrove plants.

METHODOLOGY

Study site and mangrove plant species

Koggala lagoon is one of the important mangrove forests in South of Sri Lanka, with many faunal and floral species. Therefore, mangrove plant species were collected from Koggala lagoon. Plants were collected from randomly selected three sites of the riverine mangrove forest. Plants were collected along the 100 m line transect beginning from river margin. Five mangrove plants *(Sonneratia caseolaris, Avicennia marina, Rhizophora apiculata, Acrostichum auruem* and *Bruguiera gymnorrhiza*) and two mangrove associates (*Acanthus illicifolius* and *Clerodendron inerrme)* were used.

Quantification of percentage colonization of arbuscular mycorrhizae

Freshly collected root samples were stained using a standard procedure (Phillips and Hayman 1970). The presence of arbuscules, vesicles and aseptate hyphae were used to designate AMF colonization (Brundrett *et al.*, 1984). Quantification of percentage colonization of AMF was done by the modified grid transects method (McGonigle *et al.* 1990) observing under the compound microscope at x400 magnification.

Determination of leaf free proline content

0.5 g of younger, fully expended leaves were harvested from above seven mangrove species. Leaves were ground in 5 ml of 3% sulphosalycylic acid. 2 ml of this filtrate was mixed with 2 ml of glacial acetic acid and 2 ml of acid ninhydrin. This mixture was heated in a water bath at 100 ^{0}C for 1 hour and the reaction mixture was mixed with 4 ml of toluene after cooling. Toluene part was separated after mixing and absorbance was read against toluene blank at 520 μm in spectrophotometer (Bates *et al.* 1973).

Determination of soil pH and electrical conductivity

Soil suspensions were used to measure the soil pH and electrical conductivity.

RESULTS AND DISCUSSION

Biosphere contains various ecosystems. Faunal and floral component of these ecosystems have various adaptations. Mangroves are specific ecosystems adapted to thrive in salted water. Therefore mangrove plants have adaptations of salt tolerance. Koggala lagoon is a riverine mangrove forest. So the electrical conductivity of water was low, ranged as 0.95- 4.48 ms/cm. *Sonneratia caseolaris* is preferred to grow in low salted area. *S. caseolaris* abundance was higher in this lagoon.

According to the result of Two-way ANOVA test p-value was less than 0.05. So salinity levels in root rhizosphere of seven plants were not equal at 95% confidence interval (Figure 1). *A. marina, R. apiculata* and *B. gymnorrhiza* were given values for salinity without a significant difference ($P>$ 0.05). These plants occur at river margin and they have higher electrical conductivity as 4.48, 4.53 and 4.32 ms/cm respectively. There were no *Sonneratia caseolaris* plants in salted basin area in this study site. Electrical conductivity of *Sonneratia caseolaris* root rhizosphere was very low (0.95). Statistical analysis revealed

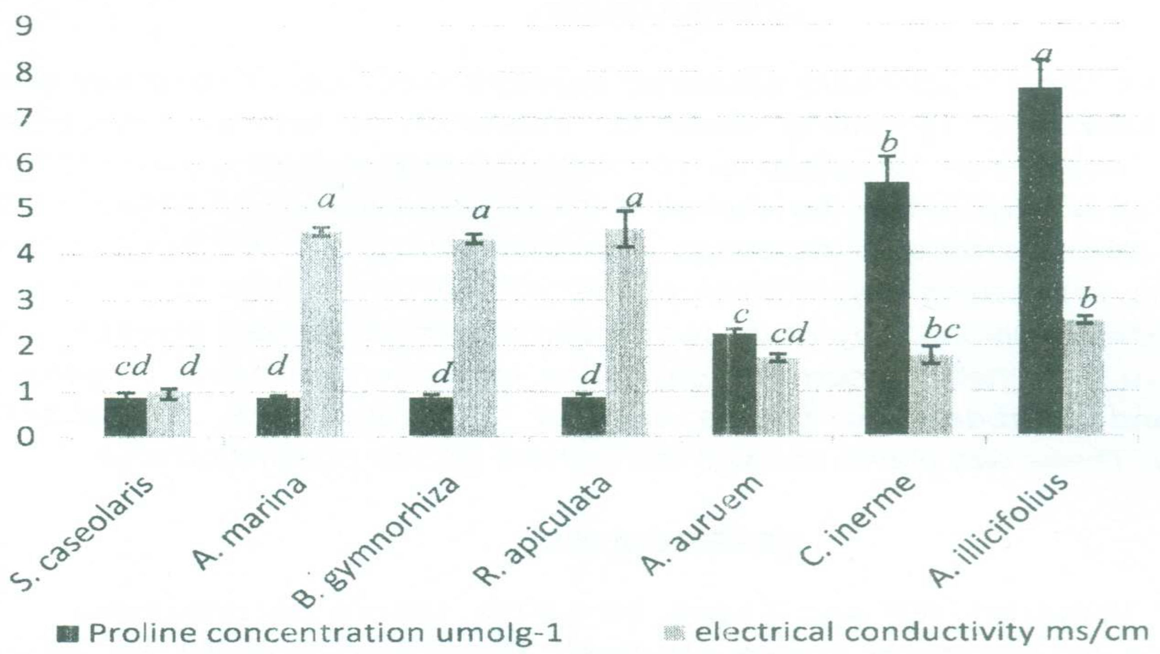

Figure 1- Variations of soil electrical conductivity and proline content (Same letter denoted non significant difference. P value is less than 0.05.)

Also there were two mangrove associative plants *Clerodendron inerme* and *Acanthus illicifolius* occur in inland area of these study sites. Rhizosphere electrical conductivity of these plants is also low (1.78, 2.54 ms/cm), but proline production was high as 5.54 and 7.62 ms/cm. *Avicennia marina, Rhizophora apiculata* and *Bruguiera gymnorrhiza* grow at higher soil electrical conductivity (4.48, 4.53 and 4.32 ms/cm). However, leaf proline concentrations were low as 0.860-0.868 µmol/g. They have salt secreted glands as a physiological adaptation and because of that there may be minimum proline requirement. Thick leaves of *Rhizophora apiculata* and *Bruguiera gymnorrhiza* can store substantial amount of water, hence there was no higher concentration of praline accumulated in cells even in high salinity condition.

According to the result of Two-way ANOVA test p-value was less than 0.05, so it can be concluded that proline levels within seven plants were not equal at 95% confidence interval. As well as there was a significant difference between *Acanthus ilicifolius* and other six plants. *Clerodendron inerme* also showed the high proline production (5.54 µmol/g) which was significantly different from *Acanthus ilicifolius.* There was no difference of proline with *Avicennia marina, Rhizophora apiculata, Bruguiera gymnorhiza* and *Sonneratia caseolaris.*

CONCLUSION

There was no clear relationship observed between electrical conductivity and proline concentration. It was a result of interaction of soil pH, electrical conductivity and other adaptations of plant. Leaf proline concentration increases with acidity in soil. As well as if there were high acidity, arbuscular mycorrhizal colonization also increases. *Acanthus illicifolius* and *Clerodendron inerme* were arbuscular mycorrhizal plants. Others were non mycorrhizal. Higher salinity conditions may lower the mycorrhizal colonization. Hyphae and vesicles were prominent mycorrhizal structures in above two plants. *Acanthus illicifolius* and *Clerodendron inerme* showed 35% and 30% colonization respectively. These two plants showed the highest proline production also.

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