Salinity Tolerance of Aegiceras corniculatum and Ceriops tagal in the coastal area of Karachi, Pakistan

Muhammad Ayyaz1, Jafron Wasiq1, Fuad Muhammad1, Waqar Ahmed3, Muhammad Yaseen2, Maria Ashraf1, Muhammad Abdul Rahman4

1Graduate Program of Environmental Science, School of Postgraduate Studies, Universitas Diponegoro, Semarang Indonesia
2College of Ecology and Environment, Hainan University, Haikou 570228, China; 20181112909153@hainanu.edu.cn
3Institute of environmental studies, University of Karachi, Karachi, 75270, Pakistan
4Department of Botany, University of Okara, Okara 53600, Pakistan

*Correspondence: ayazhussain963@gmail.com

Abstract. Indus delta with its coastal zones is the most significant coastal environment in Pakistan for mangroves and associated ecosystems. Mangrove forests are important components for maintenance of ecosystems in severe environments. The purpose of the study is to assess the mangrove species’ tolerance to salinity. Aegiceras corniculatum and Ceriops tagal (C. tagal) propagules were grown in pots with sandy soil and subirrigated with 25, and 50% nitrogen-fortified saltwater for this research. The propagules were procured from the Indus delta. We raised seedlings for six months as an experiment. Aegiceras corniculatum species behaved moderately in the greenhouse, whereas Ceriops tagal’s growth maximum at a salinity of 50% seawater and declined as the salinity increased. Furthermore, Ceriops tagal a non-secretor, accumulated more sodium and chloride ions while severely restricting the availability of other ions. This species might therefore be used to repair intertidal ecosystems, which frequently get freshwater. During a six-month the experimental cultivation period, and measurements were taken of the seedlings’ length, weight at planting, and number of leaves. Maximum growth was observed in 50% seawater, and as the salinity increased, it became worse. Three times a week, fresh water was provided to wash away the excess salt. It has been discovered that medium and large-sized propagules function better in a greenhouse environment than small-sized ones. In order for plant seeds to successfully reproduce, the environmental conditions in which they disseminate and settle must be suitable for them. For establishment and dissemination inside the greenhouse environment, the propagules of viviparous mangrove species appear optimal.

Keywords: Indus Delta; Aegiceras Corniculatum; Ceriops tagal; Salinity; Propagules; Mangroves; Ions; water relations

1. Introduction

Mangroves are located in the Indus Delta in the province of Sindh, which covers an area of around 600,000 hectares and stretches from Sir Creek in the south to Korangi Creek in the north. The Indus delta’s mangroves are exceptional because they are the biggest dry climate mangroves in the world (Amjad et al. 2007), (Hilal et al. 2019), demonstrated that mangrove forests are made up of tropical trees and plants that resemble woody shrubs that grow in the intertidal zone and support a rich ecology. A broad range of flora and fauna species may be found in the mangroves, one of the most biologically varied coastal ecosystems. Mangroves are also rich in organic matter and nutrients (Pawar 2013). Abbas et al. 2012, revealed that mangrove forests are made up of halophytic trees and shrubs that occur in tropical and subtropical parts of the world and are a source of blue carbon and ecological diversity (GMA 2021).

Eight various mangrove species may have formerly existed in the Indus Delta. However, there are currently just four species remaining. The species makes up over 95% of the mangroves in the Indus Delta (Mukhtar et al. 2012). Near the Indus River’s discharge at Keti Bunder, there are small areas of and (Amjad et al. 2007). Aegiceras corniculatum is a
small tree or shrub, and a kind of mangrove that grows along tropical and subtropical coastlines. Mangrove forest species play an important role in medical benefits. The local population living close to the seaside uses extracts of that are obtained from the stems as anti-inflammatory, anti-rheumatic, anti-diabetic, and anti-asthmatic treatments (Bandaranayake, 1998, 2002). Aziz and Khan (2001) claimed that Ceriops tagal in Pakistan shown optimum growth at a salinity of 50% seawater and deteriorated with increasing salinity. Data on water relations revealed that this species behaved as an Osmo conformer to changes in external water potential by gradually altering its internal water potential.

The mangrove forests play an important protective and productive role for the economy’s growth as well as for the biodiversity of free-living, crawling, borrowing, sessile, climbing, and other organisms (Rehman et al. 2018). Despite their importance, mangroves have been seriously damaged and converted to various forms of land use worldwide (Alongi 2002; Duke et al. 2007; Giri et al. 2011).

Mangrove trees help to cover the coastal margins in order to protect the land and environment from natural catastrophes such as tsunamis and earthquakes (Cornforth et al. 2013; Giri et al. 2007; Porwal et al. 2012; Satyanarayana et al. 2011). The river Indus, which flows through the delta before reaching the Arabian Sea, is a major factor in the survival of these forests since it provides them with consistent freshwater supplies. The Sindh Forest Department is in charge of the 344,845 hectares of the Indus delta that have been designated as protected forests (Amjad et al. 2007).

Salinity is one of the main factors influencing the growth, spread, and productivity of mangroves (Ball & Marilyn 2002). Studies of mangrove restoration projects at various sites have shown that it affects the survival and growth of planted mangrove seedlings, and it frequently exhibits high spatial and temporal fluctuations, driven by inputs of fresh water and sea water, inundation, ground water seepage, and evaporation (Bosire et al. 2008; Kirui et al. 2008; Krauss et al. 2008; Hoppe-Speer et al. 2011). Due to their halophytic nature, mature mangrove species are salt tolerant as well as adjusted to difficult situations in the environment, such as variation in salinity and soil pH (Parida & Jha 2010; Krauss & Ball 2013).

Low water potentials, ion toxicities, nutrient deficits, or a combination of all these variables may all affect plant development in high medium salinity environments (Khan et al. 2000). Young, recently planted mangrove species, however, may be particularly susceptible to changes in the physical characteristics of the water and soil, such as soil pH and salinity (Wakushima et al. 1994; Joshi & Ghose 2003). The range of salinities that mangrove plants may survive in the wild varies by species and is based on their capacity to maintain a high-water usage efficiency (Reef & Lovelock 2015). The present study was designed to investigate the relative salt tolerance of two mangrove species Aegiceras corniculatum and Ceriops tagal with artificial salinity variation of 25% and 50% at Indus delta Keti Bandar, Pakistan.

2. Method

2.1. Study area

This survey was conducted for the study of mangrove species of and of Indus delta. The Indus delta area is situated between 67o 50’ and 68o 15’ East latitude and 25o 50’ to 24o 50’ North latitude. It was designed by the Indus River’s activity, which has its source in the Himalayas. The primary spillway is located near Keti Bunder. The river’s mouth was once quite near to Karachi, but due to geological processes, it was moved from the rocky western Kohistan tract boundaries to the sandy eastern portions. As a result, the region may be classified as desert. The velocity of wind during these months’ ranges from 12 km/h to 35 km/h. This research was based on first-hand observation. Other species were available, but just two of them were chosen for the research of Primulaceae and Rhizophoraceae.

Figure 1. Keti bander, Thatta, Sindh, Pakistan; https://goo.gl/maps/ARmGBjPus4YadbJP8
2.2 Survey of Local Communities around Indus Delta

The mangrove forest along the Indus delta, keti bandar Karachi, Pakistan, was surveyed in November 2022. The main objective of this survey was to collect additional information about the mangrove forest from the local communities. In order to collect all the information regarding mangrove forests, we first arranged meeting with Muhammad Khan Jamali, an officer of the mangrove forest department. After meeting with the forest department personnel. We visited the field area. We encountered more than 50 peoples some queries about mangrove forest species. When survey completed, the officer from the forest department scheduled a meeting with us to discuss mangrove awareness for local communities. A speech on the significance of mangrove forest species and the effects of deforestation was also given by my colleague Maria Ashraf.

2.3 Collection of Propagules of Mangrove species

In the months of July and August 2022, the mature mangrove propagules of the species Aegiceras corniculatum and Ceriops tagal were collected at Keti Bunder in the Indus Delta (24°22′14.25″N, 67°26′38.41″E). Propagules were stored in moist gunny bags after collection for no more than 3–4 days in the refrigerator to prevent water loss owing to evaporation (Rasool & Saifullah, 2000b). The propagules of Aegiceras corniculatum and Ceriops tagal are normally feasible in the local environment for about a week without any pre-treatment. As a result, Rasool and Saifullah (2000b) reported that pre-treating propagules have always resulted in better results than leaving them untreated.

2.4 Method for Nursery Techniques of Aegiceras corniculatum and Ceriops tagal

and propagules were collected using this method. Propagules turn from light green to dark green when seeds ripen. Late August and early September are the optimum times to harvest them since they are more numerous and healthier (Rasool & Saifullah, 2000b). They were available from August to October of each year. They were partially hide in the plastic bags packed with dirt. In polythene bags, germination starts two weeks after planting and has a 60% success rate.

2.5 Sowing Propagules in the Polythene Bags:

The soil for these bags were come from the Institute of Environmental Studies University of Karachi, and the polythene bags were 5 x 10 inches in size with 4-6 holes in the center and edges of the bags for aeration and drainage of excess water. Before sowing, soil-filled polythene bags with fresh water were supplied, and the weight and length of the propagules were measured for initial research. Then, the propagules were sown in the polythene bags. As soon as they arrive, seeds were unpacked and divided into three sizes: big size propagules (average weight: 4.32g), medium size propagules (average weight: 3.60g), and small size propagules (average weight: 3.60g). The chosen specie's propagules were placed in a polythene bag filled with 95% silty soil after initial length and weight measurements are taken.
and the bag will be monitored often. In order to eliminate excess salt from the propagules in polythene bags, nutrients such as saltwater, Hoagland solution, and fresh water are also provided to the propagules.

2.6 Preparation of solutions

Firstly, three various solutions were applied on the propagules during the spring season in March 2022. Two main solutions were applied on the propagules for instance sea or salt water on 25% concentration and 50% concentration, along with Hoagland nutrient solution was also employed. All these solutions were arranged during the lab experiment.

To make a solution of sea or salt water at a concentration of 25%, dissolve 13.5g of sea salt in 6 liters of freshwater. In addition, 55g of sea salt was dissolved in freshwater and diluted with 6 liters of water to create a 50% solution of salt water. For the preparation of Hoagland nutrient solution, in total components were added which are given below (Table 1).

<table>
<thead>
<tr>
<th>Nutrients</th>
<th>Formula</th>
<th>Mol. Wt.</th>
<th>Per liter of nutrient solution</th>
</tr>
</thead>
<tbody>
<tr>
<td>Potassium nitrate</td>
<td>KNO</td>
<td>101.1</td>
<td>5 ml of 1 M</td>
</tr>
<tr>
<td>Calcium nitrate</td>
<td>Ca (NO)₂ 4HO</td>
<td>236.15</td>
<td>5 ml of 1 M</td>
</tr>
<tr>
<td>Monopotassium phosphate</td>
<td>KHPO₄</td>
<td>136.09</td>
<td>1 ml of 1 M</td>
</tr>
<tr>
<td>Magnesium sulfate</td>
<td>MgSO₄.7HO</td>
<td>246.47</td>
<td>2 ml of 1 M</td>
</tr>
<tr>
<td>Micronutrient stock solution</td>
<td></td>
<td></td>
<td>1 ml of stock solution</td>
</tr>
<tr>
<td>Iron chelates</td>
<td>Fe-EDTA</td>
<td></td>
<td>1-5 ml of 1000 mg/l</td>
</tr>
<tr>
<td>Micronutrient stock solution</td>
<td></td>
<td></td>
<td>Per liter</td>
</tr>
<tr>
<td>Boric acid</td>
<td>H₃BO</td>
<td></td>
<td>2.86 g</td>
</tr>
<tr>
<td>Manganese chloride – 4 hydrate</td>
<td>MnCl₂.4HO</td>
<td></td>
<td>1.81 g</td>
</tr>
<tr>
<td>Zinc sulfate – 7 hydrates</td>
<td>ZnSO₄.7HO</td>
<td></td>
<td>0.22 g</td>
</tr>
<tr>
<td>Copper sulfate – 5 hydrates</td>
<td>CuSO₄.5HO</td>
<td></td>
<td>0.08 g</td>
</tr>
<tr>
<td>85% Molybdic acid</td>
<td>MoO</td>
<td></td>
<td>0.02 g</td>
</tr>
</tbody>
</table>

3. Results

Our Results revealed that Ceriops tagal Plant height was boosted at 50% seawater and reduced with a subsequent rise in salinity based on the number of leaves and plant height as shown in (Figure 2). Plants were tallest in Ceriops tagal followed by Aegiceras corniculatum. Water potential became progressively more negative in Ceriops tagal with the increase in media salinity. In comparison to Aegiceras corniculatum, Ceriops tagal showed the lowest stomatal conductivity at all salinities. Both sodium and chloride ions were accumulated in the smallest amounts by Ceriops tagal.
The growth of *Ceriops tagal* well at 25% sea water solution. This graph shows that the Plant height of the *Ceriops tagal* (Figure 1). At a 50% dilution of saltwater, *Ceriops tagal* seedlings and saplings produced the most biomass; lower and higher dilutions produced less biomass. Plants exposed to NaCl developed salt crystals on the top surface of their leaves, in contrast to control plants that did not. These crystals were made of salts produced by the plants’ salt glands. The rate of salt excretion increased linearly as salt treatment time increased as shown in (Figure 2).

Young leaves showed a proline content that markedly rose from 25% low salinity to 50% high salinity. The proline concentration had minimal impact as the salinity was increased further. Despite the proline concentration being significantly lower than in younger leaves, the trend persisted in older leaves as shown in (Figure 3). In seedlings and saplings, neither the number of nodes nor the number of leaves changed up to a 50% seawater treatment. Our results showed that when salinity increased, every type of plant we looked at dramatically reduced their stomatal conductance. Despite the fact that both factors were significant, salinity impacts frequently had a significantly bigger impact on the explained variation (more SS) than the influence of growth stage (seedlings versus saplings). The observation that their weight in relation to seedlings hasn’t altered much since *Ceriops tagal* sampling reinforced this. This species was growing slowly, and stomatal conductance was declining as shown in (Figure 4).
Aegiceras is divided into small, medium, and giant propagules according to height classes. Out of which nutrients are given to the propagules as shown in (Fig 5). The overall cation concentrations (Na+, K+, Ca2+, and Mg2+) and anion concentrations (Cl) increased along with the salinity. Total inorganic ions increased at all seawater dilutions due to elevated Na+ and Cl concentrations. Salinity increased, while calcium, K+, and magnesium concentrations decreased.

3.1 Strategies for surveying technique

Based on many queries regarding the species of mangrove forest, the questioners were selected. In the year 2022, about 50 peoples provided responses on a number of significant questions. The outcomes of the typical survey reports are listed below:

1. 35% agree
2. 25% opposed
3. 20% No comments
4. 80% Strongly agree

4. Discussion

At 50% saltwater (22,000 mg/L NaCl), the biomass output of all mangrove species was greatly increased. Our studies showed that growth stimulation at 25% seawater for Ceriops tagal and Aegiceras corniculatum. This clearly shows that Ceriops tagal, which grows on the driest part of Karachi coast were more tolerant of salinity and capable of producing greater biomass when seawater interacts with Indus river flow. Salts were emitted by salt glands identified in Aegiceras corniculatum leaves, and they then crystallised on the upper surface of the leaves of plants that had received NaCl.
Salinity lowered *Aegiceras corniculatum*’s total pool of amino acids. In contrast to numerous studies (Muthukumarasamy et al., 2000; Wang & Nil, 2000), our results show that diverse plant species do not suffer an increase in the free amino acid pool when exposed to salt.

As a consequence, the NaCl treatments on *Aegiceras corniculatum* demonstrated a reduction in the total amino acid pool but no change in the protein composition, suggesting the species’ method of salt adjustment. Proline accumulation happens often in halophytes. Being an obligate halophyte and a species that secretes salt, it is intriguing to look at how proline builds up in reaction to salinity in *Aegiceras corniculatum*. Various stresses, notably salt stress, are well known to raise the proline content in the leaves of various plants (Lee & Liu, 1999; Hernandez et al. 2000).

*Ceriops tagal* cannot create salt through its leaves, but it can filter out salts through its roots (Hegemayer, 1997). Salt-tolerant plants had a low stomatal conductance and a high water use efficiency during extreme drought and salinity stress (Sharma, 1977; Werner and Stelzer, 1990; Gordon, 1993). According to (Ball & Farquhar 1984), this decreased stomatal conductance slows down CO2 uptake and accumulation, raises xylem tension, and lowers transpiration rates. 

In this study, *Ceriops tagal* substantially reduced stomatal conductance and greatly raised xylem tension. The non-secreting species *Ceriops tagal* showed internal salt concentrations that weren’t all that different from the neighbouring Avicennia marina under hyper-saline conditions (Gordon, 1993).

*Ceriops tagal* cannot create salt through its leaves, but it can filter out salts through its roots (Hegemayer, 1997), through successfully transporting ions to the leaf’s vacuoles, moving ions beyond the leaf (perhaps through cuticular transpiration; Tomlinson, 1986), and enabling salt shedding through quick leaf turnover, non-secretors may avoid salt injury (Stewart and Ahmed, 1983).

In competitive situations with high salinity and minimal flooding, *Ceriops tagal* was salt-tolerant mangrove with the capacity to thrive (Ball, 1988; Gordon, 1993). Large internal salt concentrations may help plants flourish in conditions where soil osmotic potential is significantly lower than that of saltwater due to excessive soil salinity (Ungar, 1991).

This is due to the fact that they support the preservation of the low internal potential required for water absorption. Our study found that the salt and chloride content of *Ceriops tagal* remained high and increased with salinity. As they adjust to low osmotic potentials, eukaryotic cells accumulate organic solutes with the least amount of disturbance to macromolecular stability and cytoplasmic enzyme activity (Stoery et al. 1977).

Salinity significantly enhanced the quantity of proline, and *Ceriops tagal* was a big collector of sodium and chloride ions. This increase, however, was insufficient to offset the enormous quantity of salt that was present in vacuoles. As salinity rose, the levels of soluble oxalate decreased, showing no function in osmoregulation. According to Popp 1 and Albert (1995), *Ceriops tagal* accumulates cyclitols together with Na+ and Cl to maintain osmotic balance. However, some animal behaviors result in the growth of mangrove propagules. Smith wrote about the crab damage to mangrove propagules (2001). Nonetheless, the authors found substantial monkey damage to the mangrove propagules. In accordance with our research, *Ceriops tagal* were only grown under the best circumstances if saltwater and enough Indus river water were combined on a regular basis.

Individuals of the *Ceriops tagal* species consume a lot of salt and chloride in order to maintain the osmotic balance in their tissues. Due to *C. tagal’s* strong resistance to salt, it is possible to grow it close to the water, where it would encounter daily floods with minimal change in the salinity of the soil.

The results of the current inquiry was only supported by a small number of applicable published studies. Under the only published study on the germination of *Aegiceras corniculatum* propagules, Siddiqi (2001) reported a 100% success rate for germination under fresh water conditions in the nursery.

Under the same conditions, this investigation discovered exactly what you would expect. However, Siddiqi (2001) did not look at the effect of rising salinities on germination. According to Saenger (2002), *Aegiceras corniculatum* is very good at rejecting salt while absorbing water, and mangroves have a salt exclusion mechanism in its roots. The plant was therefore exposed to salt during germination since the salt exclusion mechanism doesn’t function until after the beginning of roots.

### 5. Conclusion

From this study, it is concluded that Mangrove species like *Ceriops tagal* readily germinate in greenhouses or artificial environments when treated with certain procedures using saltwater at two distinct percentages of 25 and 50%, Hoagland solution, and fresh water. Salinity considerably hinders seedling development in *Aegiceras corniculatum* but not propagule germination. For *Aegiceras corniculatum* in Pakistan, salinity levels of up to 25% would be considered optimum for both germination and growth. When planting the species in highly salinized areas, it was suggested to utilise nursery-grown seedlings of the species since *Aegiceras corniculatum* seedlings can withstand higher salinities well.
Ceriops tagal displayed a gradual rise in tissue water and osmotic potentials with increasing salinity of the medium. Showed that it uses an osmoconformer approach to keep its osmotic balance. This might suggest that low water usage efficiency and low carbon absorption are to blame for Ceriops tagal slower development.

The loss of mangrove species was closely tied to the reduction in fresh water and silt supplies to the Indus Delta during the previous 50 years. The decline of mangrove species was also linked to the increase in the amount of untreated residential and industrial wastewater discharges from Karachi and its coastal zones.

Due to constant protection from foreign competition, domestic industries are noncompetitive and have inefficient manufacturing and post-production technologies and procedures. Additionally, when port channel dredging is being done, all international dredging laws are entirely disregarded. The present level of marine pollution in the Delta, which is anticipated to reach Somiani in Baluchistan and poses a constant threat to biodiversity, has impeded the establishment of mangroves.

Aknowledgment.

The authors would gratefully like to thank supervisors, department of Master of Environmental Science, Universitas Diponegoro, and University of Karachi, Pakistan for the support in this study. I also want thank Allah Almighty for giving me courage, thanks to my family, and my friends.

References


