Growth and Root Development of Four Mangrove Seedlings Under Varying Salinity

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Abstract. This present study describes four mangrove seedlings namely Bruguiera cylindrica, B. sexangula, Ceriops tagal, and Rhizophora apiculata in response to salinity with particular emphasis to root development. The seedlings of four mangroves were grown for 5 months in 0%, 0.5%, 1.5%, 2.0% and 3.0% salt concentration. Salinity significantly decreased the growth (diameter and plant height) of all mangrove seedlings. Root developments were observed from the tap and lateral root. The number, length and diameter of both roots-typed of B. cylindrica, B. sexangula and C. tagal seedlings significantly decreased with increasing salt concentration with optimum development at 0.5% salinity. By contrast, the number, length, and diameter of tap root of R. apiculata seedlings were significantly enhanced by salt with maximal stimulation at 0.5%, and this increase was attenuated by increasing salinity. On the other hand, lateral root development of R. apiculata significantly thrived up to 1.5% salinity then decreasing with the increasing salinity. The different response of root development suggested valuable information for mangrove rehabilitation in North Sumatra and their adaption to withstand salt stress.

Keywords: Main root, rehabilitation, Rhizophoraceae, salt stress, water stress.

1. Introduction

Mangroves are halophytes that thrive in the inter-tidal zone of tropical and subtropical climates. One striking feature of mangrove plants is their ability to grow in various levels of salinity, ranging from freshwater to hypersaline environments [1]. Thus, the salinity environment of mangrove habitat may vary day to day or month to month. The mangrove root hence faces transient increases and decreases in soil salinity under natural field conditions. It is important therefore to get more insight into the physiological aspects of mangrove plants to cope with salinity.

Indonesia is one of the world’s great mangrove nations, which is 22.6 percent of the global total. However, the Indonesian mangrove area has been lost from 4.2 million in 1980 to 3.1 million in 2011 [2]. Mangrove forests in North Sumatera area distributed in the east coast of Sumatera Island mainly in the Karang Gading and Langkat Timur Laut Wildlife Reserve (KGLTLWR) and are rapidly threatened due to anthropogenic activities such as replacement of mangroves to aquaculture and oil palm plantation, filling and urban development [3]. Mangroves are one of the most threatened ecosystems all over the world today due to the deforestation and degradation [4]. Conservation efforts of mangrove implicate not only to protect the coastal areas and human communities from seawater intrusion and potential changes in the sea-level rise but also to make sure the availability of mangrove resources for future use through adaptation and mitigation to shifting environments.
The response of mangrove to salinity has mainly been studied in leaves [5-7], and limited information is available on salt stress-dependent changes of root development from mangroves in North Sumatran. It is important to note that the success of rehabilitation and conservation effort in the region. Our previous studies shed light on the optimal growth that 0.5% salinity was the optimal growth for *Kandelia candel* and *Bruguiera gymnorrhiza* seedlings grown in a glass house [7]. It is important to get more insight into the morphological aspects of mangrove seedlings to cope with salinity, which may experience transient periods of exposure to high salt concentrations. The present study, therefore, aims to investigate the response four mangrove seedlings namely *Bruguiera cylindrica*, *B. sexangula*, *Ceriops tagal*, and *Rhizophora apiculata* in response to salinity with a particular reference to root development.

2. Materials and Methods

2.1. Experimental design

Mature and healthy propagules of four mangrove species namely *Bruguiera cylindrica* Blume, *B. sexangula* (Lour.) Poir., *Ceriops tagal* C. B. Rob., and *Rhizophora apiculata* BL. (Rhizophoraceae), were collected from Pulau Sembilan, Langkat, North Sumatra, Indonesia. These propagules were planted in bottle pots with sand under varying salinity with exposure to natural temperature and sunlight in a greenhouse of Faculty of Agriculture, University of Sumatera Utara, Medan, Indonesia. Mature propagules of *B. cylindrica* were green to purplish green in color, 0.5-1.0 cm diameter and 10-15 cm long; mature propagules of *B. sexangula* were green to purple-tinted brown, 1.5-2 cm diameter, 6-12 cm long. On the other hand, mature propagules of *C. tagal* were green to brown hypocotyl, yellow cotyledonary collar, 0.8-1.2 cm diameter, up to 25 cm or more long. While mature propagules of *R. apiculata* were green to brown hypocotyls and red cotyledonary collar, 20-25 cm long and 1.3-1.7 cm diameter.

An artificial seawater solution was prepared by dissolving a commercial salt powder (Tropical Marine Sea Salt, Thailand) to adjust the salinity concentrations to 0.0%, 0.5%, 1.5%, 2.0% and 3.0% (equal to sea water level) according to the manufacturer’s protocol. Each pot was irrigated with 1000 mL artificial seawater solution. The salinity in this study was defined as the mass of salt powder/weight of solution [8]. In each pot, the treatment of salt concentration was checked every week during the experiments using an S/Mill-E salinity refractometer (Atago Co. Ltd., Tokyo, Japan) and adjusted accordingly. Ten plants per treatment were grown for five months. After five months cultivation, the four mangrove seedlings were harvested, washed and the leaves and roots were immediately stored at -4 °C for further analysis.

2.2. Growth measurement

The growth of *B. cylindrica*, *B. sexangula*, *C. tagal* and *R. apiculata* seedlings under varying salt concentration was determined by the stem height and diameter of the plants. Thus, the stem heights and diameters of four mangrove species (10 seedlings each treatment) after five months of cultivation were the indices of growth in this experiment.

2.3. Root development

Root development of *B. cylindrica*, *B. sexangula*, *C. tagal* and *R. apiculata* seedlings under varying salt concentration were determined by a number of total roots, roots length and roots diameter as previously described [9]. A number roots were measured manually using counter after five months seedlings harvested. Measurement of root length was performed manually after harvesting. The diameter of root was measured after five months collected using the digital caliper. Root anatomy parameters were separated into tap root, and lateral roots as previously reported [9].

2.4. Statistical analysis

Data were analyzed by one-way analysis of variance (ANOVA) followed by Dunnett’s test for comparisons of all treatments against the control. The value of $P < 0.05$ was selected as a limit of
statistical significance. All statistical analyses were performed using the SAS 9.1 statistical software program (SAS Institute Inc. Cary, NC, USA).

3. Results and Discussion

3.1. Effect of salinity on the seedlings growth

Mangrove seedling growth was measured by the height and diameter of the plants as described in Materials and method. The growth of *B. cylindrica* decreased in the presence of salt (Fig. 1 A and B). Similarly, salt stress also significantly reduced the height and diameter of *B. sexangula* seedlings as well as *R. apiculata*. In contrast to this observation, the height of *C. tagal* seedlings thrived up to 0.5% salt concentration of both height and diameter of seedlings, and this increase then was attenuated by salinity when enhanced more than 1.5% salt concentration (Figure 1).

Mangrove species often show growth stimulation at low salinity (25% seawater/0.5% salt concentration) and intermediate salinity (50% seawater/1.5% salinity) and then a decline in growth with further elevating in salt concentration [10-12]. However, the range of salinity in which the mangrove plant can survive varies according to the species [11]. This study suggested that mangrove seedlings survived to adapt either in saline condition or fresh water.

The present study also well agreed with the previous reports on the optimal growth at 0.5% salinity in *Kandelia candel* and *Bruguiera gymnorrhiza*, respectively. The growth of both species slightly increased after removal to salinity [7]. Furthermore, the optimum growth at 1.5% salt concentration found in *Avicennia marina* and *Rhizophora stylosa* seedlings grown in a greenhouse [8].

![Figure 1](image_url)

**Figure 1.** Effect of salinity on plant height (A) and diameter (B) of *B. cylindrica* (●), *B. sexangula* (■), *C. tagal* (□), and *R. apiculata* (▲) seedlings. Data are expressed as the means ± SE (n=10) for growth measurements. The asterisk indicates a statistically significant difference from 0% at *P* < 0.05 using Dunnett’s test.

The tolerance of mangrove species to salinity, therefore, followed the order of *C. tagal* < *B. cylindrica* < *B. sexangula* < *R. apiculata*, which is by their habitat zonation in North Sumatran mangroves. Our current result, therefore, suggested important information mangrove adaptation and to the success of rehabilitation and conservation efforts in the region.

3.2. Effect of salinity on the root morphology

The root characteristic of the five-month-old seedlings of four mangrove species under varying salt concentration is summarized in Fig. 2. *B. cylindrica*, *B. sexangula*, and *R. apiculata* seedlings consist of the precise type of root, namely tap (main) and lateral root. On the other hand, only *C. tagal* seedlings were distinguishable of uncompleted root anatomy, even though they are originated the same family Rhizophoraceae (Figure 2).
**Figure 2.** Effect of salinity on root morphology and depth (cm) of six-month seedlings of four mangroves grown with exposure to natural temperature and sunlight in a glass house. Root type: 1. tap root; 2. lateral root.

In case of *C. tagal* roots, the only seedling with 0.5% salinity has been showed distinct tap root. However, it is noteworthy that leaves are present in *C. tagal* under varying salinities. It has been reported that *C. tagal* from Pakistan showed an optimal growth at 50% seawater salinity [12].

### 3.3. Effect of salinity on the root development

Knowledge of root seedling in response to salinity is essential to an understanding of regeneration pattern of mangrove. Tables 1-4 summarizes the effect of salinity on the root development of *B. cylindrica*, *B. sexangula*, *C. tagal* and *R. apiculata*. For *B. cylindrica* seedlings, the significant decrease was found only 3% salinity in parameters of some taps and lateral root, both tap, and lateral root length as well as tap and lateral root diameter (Table 1).

**Table 1.** Effect of salinity on the root development of *B. cylindrica* seedlings

| Salinity | Number of roots | Root length | Root diameter |
|----------|----------------|-------------|---------------|
|          | Tap | Lateral | Tap | Lateral | Tap | Lateral |
| 0.0%     | 10.0±0.6 | 92.6±4.3 | 6.8±0.2 | 2.7±0.3 | 2.4±0.3 | 0.2±0.0 |
| 0.5%     | 9.4±0.5 | 91.6±7.6 | 5.4±0.6 | 2.5±0.2 | 1.7±0.1 | 0.2±0.0 |
| 1.5%     | 8.7±0.4 | 87.4±6.3 | 5.2±0.6 | 2.5±0.3 | 1.7±0.1 | 0.2±0.0 |
| 2.0%     | 8.0±0.9 | 62.0±16.9 | 4.9±0.6 | 2.3±0.4 | 1.7±0.2 | 0.2±0.0 |
| 3.0%     | 2.4±0.3<sup>a</sup> | 1.8±5.3<sup>a</sup> | 2.4±0.2<sup>a</sup> | 0.2±0.1<sup>a</sup> | 1.5±0.1<sup>a</sup> | 0.1±0.0<sup>a</sup> |

Data are represented as the means ± SE (*n*=10)

<sup>a</sup>Significantly different from 0% at *P* < 0.05 using Dunnett's test

Similarly, number, length, and diameter of both roots-typed of *B. sexangula* and *C. tagal* seedlings were significantly decreased with increasing salt concentration with optimum development at 0.5% salinity (Table 2-3). By contrast, the number, length, and diameter of tap root of *R. apiculata* seedlings were significantly enhanced by salt with maximal stimulation at 0.5%, and this increase was...
attenuated by increasing salinity. On the other hand, lateral root development of *R. apiculata* significantly thrived up to 1.5% salinity then decreasing with the increasing salinity (Table 4).

**Table 2.** Effect of salinity on the root development of *B. sexangula* seedlings

| Salinity | Number of roots | Root length | Root diameter |
|----------|-----------------|-------------|---------------|
|          | Tap Lateral     | Tap Lateral | Tap Lateral   |
| 0.0%     | 3.5±0.4 61.5±5.7 | 12.7±0.8 4.2±0.2 | 3.8±0.3 0.5±0.1 |
| 0.5%     | 4.3±0.5 68.0±6.4 | 14.1±1.0 4.2±0.2 | 3.7±0.3 0.6±0.1 |
| 1.5%     | 3.9±0.4 50.8±4.6 | 12.4±6.3 3.5±0.2 | 2.8±0.3a 0.5±0.1 |
| 2.0%     | 3.1±0.6 19.8±2.2 | 4.5±0.4 2.8±0.4 | 2.2±0.2a 0.6±0.1 |
| 3.0%     | 1.4±0.4a 1.9±0.9a | 0.9±0.2a 0.4±0.2a | 1.2±0.2a 0.2±0.1a |

Data are represented as the means ± SE (*n*=10)
aSignificantly different from 0% at *P* < 0.05 using Dunnett’s test

**Table 3.** Effect of salinity on the root development of *C. tagal* seedlings

| Salinity | Number of roots | Root length | Root diameter |
|----------|-----------------|-------------|---------------|
|          |                |             |               |
| 0.0%     | 0.0±0.0        | 0.0±0.0     | 0.0±0.0       |
| 0.5%     | 3.2±0.5        | 1.9±0.2a    | 1.2±0.1a      |
| 1.5%     | 3.6±0.9        | 0.8±0.3     | 0.2±0.1       |
| 2.0%     | 5.2±0.6        | 1.2±0.1     | 1.0±0.1       |
| 3.0%     | 2.8±1.0        | 0.5±0.2     | 0.2±0.1       |

Data are represented as the means ± SE (*n*=10)
aSignificantly different from 0% at *P* < 0.05 using Dunnett’s test

**Table 4.** Effect of salinity on the root development of *R. apiculata* seedlings

| Salinity | Number of roots | Root length | Root diameter |
|----------|-----------------|-------------|---------------|
|          | Tap Lateral     | Tap Lateral | Tap Lateral   |
| 0.0%     | 1.1±0.1 39.9±1.6 | 3.7±0.4 2.1±0.1 | 3.3±0.3 1.4±0.1 |
| 0.5%     | 1.8±0.2 46.6±2.9a | 5.3±0.6a 2.4±0.1 | 4.4±0.4 1.3±0.1 |
| 1.5%     | 2.3±0.4 56.1±3.2a | 3.3±0.3 2.7±0.1a | 3.6±0.4 1.3±0.1 |
| 2.0%     | 2.3±0.3 37.8±2.0 | 5.2±0.4a 3.3±0.1a | 3.0±0.1 1.4±0.1 |
| 3.0%     | 1.2±0.1 18.9±1.7a | 4.2±0.3 2.7±0.2a | 2.8±0.2a 1.5±0.1a |

Data are represented as the means ± SE (*n*=10)
aSignificantly different from 0% at *P* < 0.05 using Dunnett’s test

Root tissues are in contact with salt and are the primary site involved in the perception of salinity stress; they are assumed to have remarkable ability to control their Na⁺ dan Cl⁻ content [7]. When different species compared, the root development declined under varying salinity in the order of *C. tagal > R. apiculata > B. sexangula > B. cylindrica*. The different response of root growth among mangrove species suggested the critical study of salt tolerance mechanism of individual genera or species [13].
4. Conclusion

The different response of seedling growth, as well as root development from four mangrove seedlings, suggested valuable information for mangrove rehabilitation in North Sumatra and their adaption to withstand salt stress. Our research indicated the critical study of salt tolerance mechanism of individual genera or species.

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