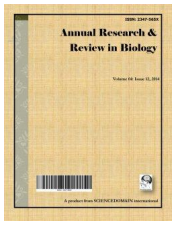

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Nursery Techniques and Primary Growth of *Rhizophora apiculata* Plantation in Coastal Area, Central Vietnam

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Authors' contributions

This work was carried out in collaboration between all authors. Author TVD designed the study, wrote the protocol and interpreted the data. Authors PND and NTT anchored the field study, gathered the initial data and performed preliminary data analysis. Authors TVD and OK managed the literature searches and produced the initial draft. All authors read and approved the final manuscript

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ABSTRACT

There are more than 3,200 km shoreline and 3,000 islands with more than 408,000 ha of mangrove forests in Vietnam. A total of 77 mangrove species were found in Vietnam including *Rhizophora mucronata, R. apiculata, R. stylosa, Kandelia candel, Avicennia alba, Sonneratia caseolaris*. The mangrove forests have been dramatically reduced because of increasing in population, food demand, aquaculture development, and urbanization. In this study, research on techniques for producing seedlings and establishing *Rhizophora apiculata* Bl. plantation was conducted in coastal area, central Vietnam. Results indicated that to produce healthy seedlings 50% length of fruit shoot must be fixed to plastic bags and 10‰ salinity water must be used for daily watering. In addition, 5-month-old seedlings must be sea-watered for a duration of at least one month at nursery before planting. *R. apiculata* is an opportunistic invader in coastal area central Vietnam, since it can grow in new mudflats. In low tidal areas as tide happens 100 – 299 days/year with water depth of 0.2 – 0.3 m in 4 – 8 hours/day, 36-month-old planted trees achieved 187.7 cm tall, 4.2 cm stump

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diameter, and 41.8 prop-roots per tree. It is recommended that selecting suitable mangrove tree species for planting in new mudflats is important for the success of mangrove afforestation program.

Keywords: Mangrove; mudflat; Rhizophora apiculata Bl.; seedling; tidal regime.

1. INTRODUCTION

The widespread loss and degradation of mangrove forests throughout Southeast Asia and over the world in the past decades have been well documented [1-4]. Land conversion for aquaculture is known as one of the main causes, as a result of pressures from increasing population, food production, and industrial and urban development. In addition, the mangrove ecosystem faces serious threats from global environmental change especially rapid sea-level rise [5]. Recently, the awareness regarding the ecological and economic importance of mangrove forests has grown such as shoreline protection, sediment and nutrient trapping, fish attracting [6], and belowground carbon sequestration against climate change [5,7]. Therefore, countries have engaged in the conservation of mangrove environment and restoration forestry [2,8-10]. Not only governments, but also local communities and NGOs are involving in mangrove forest development, mainly for the purposes of conservation, promoting aquaculture, and shoreline protection. NGOs are responsible for technique guidelines, while government is responsible for planning, management and financial support, and local people conduct planting, tending and protection accompanied with aquaculture development.

The objectives of growing mangrove forest vary widely from case to case as commercial forestry purpose, restoring fishery habitat, shoreline protection [11]. For example, the choice of species for commercial forestry purpose has been limited to some commercially valuable species (e.g. *Rhizophora* spp.), without considering other aspects as ecological and social-economic importance of mangrove forests. These have been leading to degraded functions of mangrove forests, epically in developing countries [11-13].

Vietnam locates in Southeast Asian region with more than 3,200 km shoreline and 3,000 islands [14]. There was 408,500 ha of mangrove forests in 1943, reducing to 290,000 ha in 1962, to 252,000 ha in 1982, and to 156,600 ha in 1999 [15]. A number of reasons were responded for such as wars, aquaculture development, and shoreline encroachment. There is a total of 77 tree species belonging to 28 families found in mangrove forests in Vietnam [16]. Some main mangrove tree species (*Rhizophora mucronata, R. apiculata, R. stylosa, Kandelia candel, Avicennia alba,* and *Sonneratia caseolaris*) have been widely used in Vietnam for purposes of rehabilitation of degraded areas, sea dike protection, and mixed shrimp farming-mangrove areas [17,18]. However, technical problems have been still left behind such as nursery, forest establishment *etc*. [18].

The objectives of this study were to search for suitable techniques in producing seedlings, and to evaluate the primary growth of 36-month-old plantation of *Rhizophora apiculata* Bl. planted in intertidal new mudflats, coastal area central Vietnam.

2. MATERIALS AND METHODS

2.1Study Site Description

This study was conducted in Thua Thien Hue Province (TTH; 16º44'30"N, 107º23'48"E–15º59'30"N, 107º41'52"E) central Vietnam (Fig. 1). TTH has a total land area of 5,033.6 km^2 with a total of 120 km shoreline and 24,000 ha of mudflats for mangrove forest development [19]. Mean annual temperature is 25.2ºC, while maximum and minimum temperatures are 42ºC and 8ºC, respectively. Mean annual rainfall is 2,995 mm. There were some years with total rainfall of up to 4,900 mm, leading to serious flooding and destroying mangrove forests [19]. There are six main rivers flowing from west to east and discharging to Vietnam East Sea, creating mudflats and lagoons for mangrove and aquaculture development.

2.2 Description of Study Species

Species *Rhizophora apiculata* Bl. was selected in this study, since it is a multi-purpose species. Wood is widely used locally. While it is known as a best species for shoreline protection in TTH and an attracting species for marine fauna.

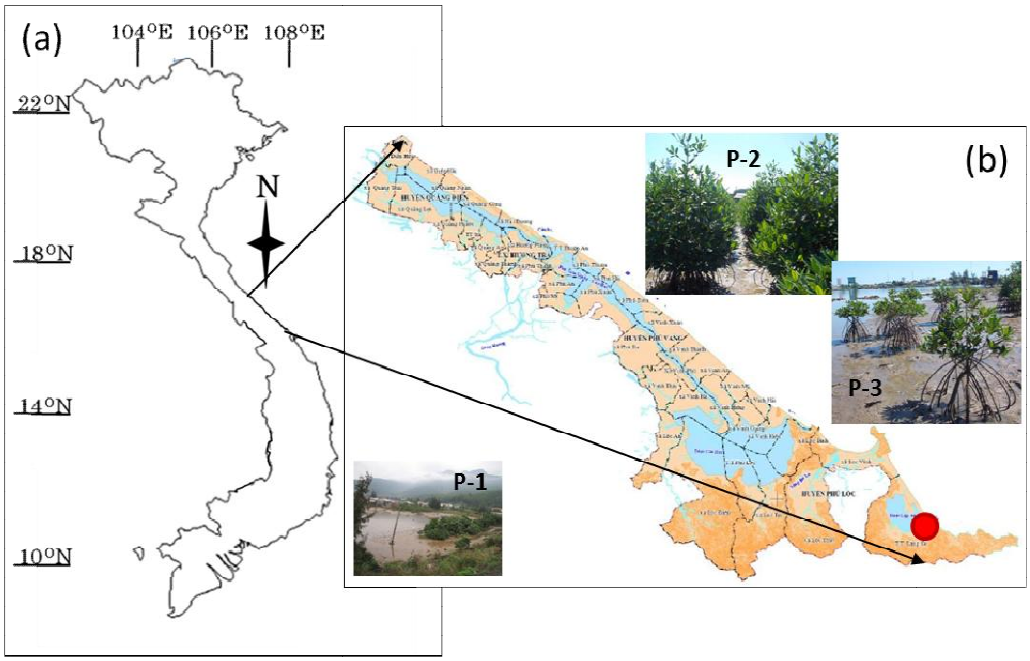

Fig. 1. Map of Vietnam (a), Thua Thien Hue Province (b), and a red dot indicates research site at Lang Co Town. P – 1 indicates experimental site for planting *Rhizophora apiculata*; P – 2 indicates 36-month-old trees in low tide area, where tide happens 100 – 299 days/year with water depth of 0.2 – 0.3 m in 4 – 8 hours/day; and P – 3 indicates 36-month-old trees in high tide area, where tide happens 300 – 364 days/year with water depth of 0.4 – 0.5 m in 6 – 10 hours/day. Photos were taken at the time of tide down in early morning

R. apiculata is an evergreen tree, native to Vietnam. It can grow up to 40 cm diameter and 30 m tall, usually reaching 8 – 10 m tall in intertidal wetland areas with variable rainfall. Fruits become ripen in August – September, when its cover turns to smooth brown (Fig. 2a). Fallen fruits are collected at local (Fig. 2a), then are sown immediately to plastic bags (Fig. 2b). Fruits (Fig. 2a) of 30 cm long, 1.2 – 1.5 cm diameter, and 30 g/fruit are widely used for producing seedlings. *R. apiculata* adapts to a wide range of soil but thrives best in fine mud stable sediments. It has been widely used for soil stabilization, shoreline protection, and wildlife/marine habitats. Its main products are timber, fuel wood, and charcoal. *R. apiculata* is a ready colonizer of new mudflat, making it opportunistically invasive [20].

2.3 Experiments on Producing Seedling

Fruits were collected from natural forests at research site (Fig. 2) and experiments were conducted locally. Medium filled into plastic bags (20 cm length and 12 cm diameter) included 1% phosphonate, 4% organic fertilizer, 25% muck (self-made by mixing 30% animal dung and 70% vegetation, then fermented), and 70% mud soil in volume. Shading 50% sunlight was conducted in the first 45 days after sowing by using black mesh tarp.

2.3.1 Effects of salinity

There were seven treatments of salinity used as 0, 5, 10, 15, 20, 25, 30‰. Fruits were sown in plastic bags with the depth of 50% fruit shoot. Each treatment included 60 seedlings with tri-replications. Experiment was arranged in randomly-completed block. Totally 420 seedlings were used for this experiment. Generally, sea water in study site has salinity of from 30‰ to 33‰. It was then mixed with an amount of freshwater for desired salinity. Watering seedlings was conducted 1 time a day at late afternoon (around 5 PM). Fertilizing and other tending activities were not applied.

2.3.2 Effects of shoot depth

The depth of fruit shoot fixed into soil at sowing was concerned in this experiment. There were

four treatments used as 50%, 33%, 25%, and 0% fruit shoot fixed to soil. Fruits with similar size of around 30 g/fruit in mass and 32 cm in length were sown in plastic bags. Each treatment included 60 seedlings with tri-replications (each replication of 20 seedlings). Experiment was arranged in randomly-completed block. Totally, 240 seedlings were used for this experiment. Sea water was used for watering one a day at late afternoon (around 5 PM). Fertilizing and other tending activities were not applied.

2.4 Experiment on Planting *R. apiculata*

This experiment was conducted at Lang Co Town (Fig. 1). There were two intertidal regimes included as (1) tide happens 300 – 364 days/year with water depth of 0.4 – 0.5 m in 6 – 10 hours/day named as high tide area (HT), and (2) tide happens 100 – 299 days/year with water depth of 0.2 – 0.3 m in 4 – 8 hours/day named as low tide area (LT). These two sites are new mudflats, where mangrove forest have never grown.

Six-month-old seedlings of 40 cm tall with 6 – 8 leaves were used in this experiment. Seedlings were produced in plastic bags at local by using sea water for watering. Planting density was 7,000 seedlings/ha (spacing of 1.5 m x 1 m, which has been widely used for mangrove forest establishment for any purposes in Vietnam). Tri-replications were used for each tidal regime, resulting in 6 replications. One hundred seedlings were used in each replication. Totally, 600 seedlings were used in this experiment. Tending activities included fixing planted trees against falling, removing materials and sea weeds from attaching to stems. These activities were conducted in the first two years after planting. Because of tidal regime, generally after planting trees in HT area were fully flooded 0.5 – 3 hours/day, while it was 0.3 – 1.5 hours/day in LT area. After six months planting, planted trees have not been fully flooded.

2.5 Data Collection

For experiments on producing seedlings, data were collected at one after sowing 6 months. All seedlings in experiment were measured. Since 6-month-old seedlings are ready to plant to the field. Data were collected for replication separately including survival rate, height (H), stump diameter (D_o), total leaves grown, and total leaves shed. For planting experiment, data were collected at 6, 12, 24, and 36 months after planting, including survival rate, H, D_o, and number of pro-roots. All survival trees were measured.

2.6 Data Analysis

Mean and its standard error were used for all parameters. Meanwhile, comparison of means of parameters among treatments for experiments on producing seedlings was assessed by univariate analysis of variance (ANOVA) and post-hoc test comparison. *T-test* was used for comparison of mean between HT and LT areas in experiment on planting *R. apiculata*. Data analyses were conducted by using SAS 9.2 (SAS Institute Inc., Cary, NC, USA).

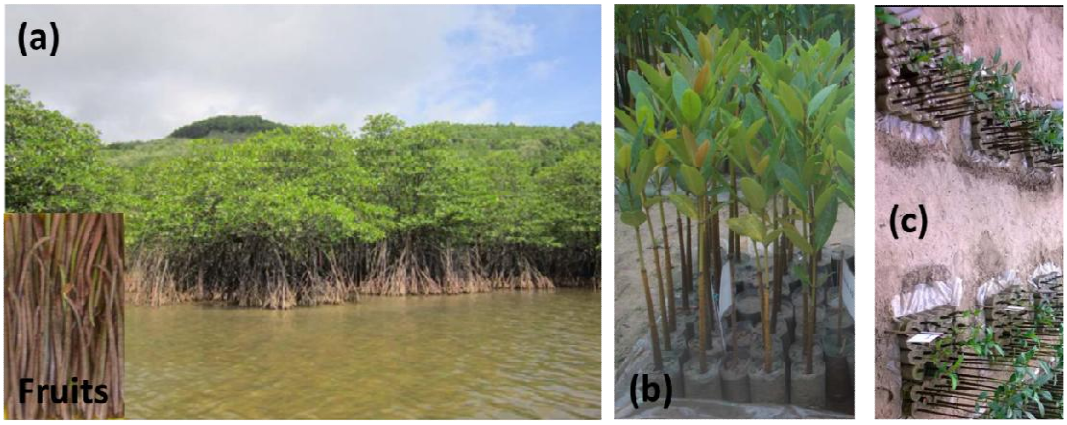

Fig. 2. Natural forest of *Rhizophora apiculata* for fruit collection (a), ready seedlings for planting (b), and experiment layout on producing seedlings (c)

3. RESULTS

3.1 Experiment for Producing Seedling

3.1.1 Effects of salinity

There was no significant difference in term of survival rate, stump diameter (D_o), total leaves shed among seven treatments of salinity (*P*> 0.05). But the difference of means of height (H) and total leaves grown was significant ($F_{3, 211}$> 10, *P*< 0.001). It was the best in the treatment of 10‰ salinity (Table 1), where seedlings achieved 26.7 cm tall and 8.3 leaves per seedling at the age of 6 month old.

3.1.2 Effects of shoot depth

There was no significant difference ($F_{3, 8}$ = 3.5, *P*> 0.05) of survival rate among four treatments (Table 2). The highest survival rate of 97.8% belonged to treatment of fixing 25% fruit shoot to soil, followed by 33% and 50% of the same survival rate of 93.3%, then lowest belonged to not fixing fruit shoot to soil. There was significant difference ($F_{3, 211}$> 10, *P*< 0.001) of H among treatments (Table 2), which was highest in 50% treatment, followed by 33%, 25%, and 0% fixing. Meanwhile, significant difference of D_o among treatments was not found. Total leaves grown were significantly different among treatment ($F_{3, 211}$> 10, *P*< 0.001), but the difference between treatment of 50% fixing and 33% fixing was not significant (Table 2). Total leaves shed were also significantly different among treatment ($F_{3, 211}$> 10, *P*< 0.001).

3.1.3 Experiment on planting *R. apiculata*

There were no significant differences (*P*> 0.05) in term of survival rate, H, D_o, and number of prop-roots after planting 6, 12, and 24 months between low tide area (LT) and high tide area (HT; Fig. 3). But it was significantly different after planting 36 months (Fig. 3). *R. apiculata* grew better in LT area than that in HT area. At 36-month-old, *R. apiculata* achieved 187.7 cm tall, 4.2 cm D_o, and 41.8 prop-roots per tree in LT area, compared to 133.2 cm tall, 2.5 cm D_o, and 22.0 prop-roots in HT area (Figs. 3b, c, d). Survival rate of *R. apiculata* at 36-month-old was 93.3% in LT area and 74.4% in HT area (Fig. 3a).

4. DISCUSSION

4.1 Nursery Technique

There was no significant difference of survival rate of 6-month-old seedlings among treatments on depth of fruit shoot fixed. It is consistent with study for other mangrove tree species such as *Sonneratia caseolaris, Kandelia candel* [21,22]. However, time required to final germination was significantly different [18]. Pham [18] indicated that it takes less than 30 days to final germination if fruit shoot was fixed to the soil regardless of depth, comparing to more than 40 days without fixing fruit shoot to soil. In nature, fruits of other mangrove tree species can float more than 50 days before germinating [22]. Fruit shoot depth fixed to the soil much affected on growth of 6-month-old seedlings (Table 2). The deeper fruit shoot fixed the better growth of seedlings is. The same pattern for total leaves grown. These may result from absorbing more nutrients from deeper root system of seedlings in fixing 50% fruit shoot treatment, compared to others (Table 2). Meanwhile, there have no clear explanations for high number of leaves shed in 50% treatment compared to others (Table 2). Nguyen and Phan [23] indicated that *R. apiculata* peaks leaf shedding in winter when temperature falls down to less than 15ºC. While, this research was conducted in Thua Thien Hue Province with minimum temperature of 10ºC in winter. But this should happens to all treatments other than only treatment of 50% fruit shoot fixed.

R. apiculata is a species native to intertidal mudflats with variable salinity [20]. Obviously, using freshwater for watering seedlings is not a good choice. Even, others [16,24] indicated that freshwater is best for germination rate and shortening germination duration. The best survival rate and growth of 6-month-old seedlings belonged to treatment of using 10‰ salinity for watering (Table 1). It is obvious that seedlings are still weak to against harsh environment as salinity. Therefore, low salinity will make seedlings easier in absorbing water and nutrient from soil to sustain their growth. While, too much lower salinity in water will lose salinity in cells, leading to abnormal growth (Table 1). Even using 10‰ salinity resulted in best growth and survival rate of 6-month-old seedlings, it may not be a good choice to plant them directly to the field because of low salinity content in its cells compared to 33‰ salinity in seawater. It suggests that seawater must be used to water these seedlings for a duration of at least a month, making them adapt to salinity of seawater. The experiments on effects of salinity and fruit shoot depth fixed to soil may partly explain for less success of natural regeneration of mangrove tree species [3,22], as fallen fruits are not easily to fix to soil from tidal regime and high salinity of seawater of 33‰ [18] compared to the best salinity 10‰ used in this study.

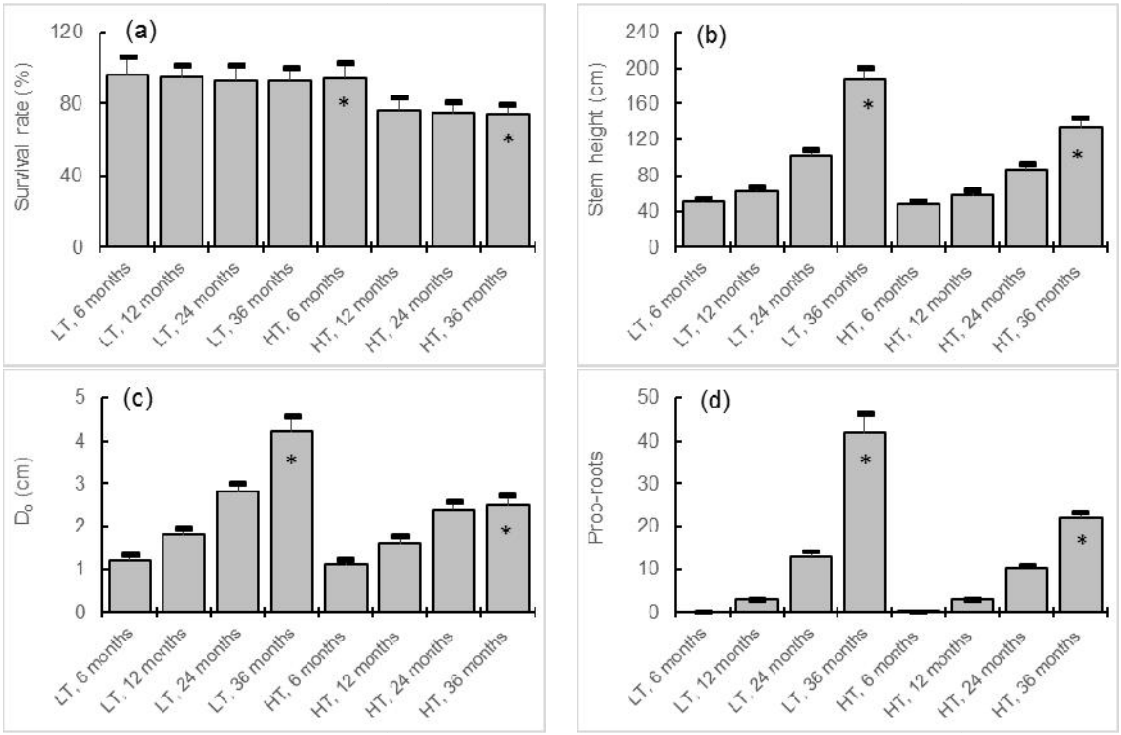

Fig. 3. Survival rate (a), growth of stem height (b), growth of stump diameter/ D_o(c), and number of prop-roots (d) of 36-month-old *Rhizophora apiculata* plantation. Bars indicate mean + standard error. Bars with asteristes indicate the difference of means between low tide (LT) and high tide (HT) by *t*-test at *P* = 0.05. In high tide, water covers 300 – 364 days/year with a depth of 0.4– 0.5m. In medium tide, water covers 100 – 299 days/year with a depth of 0.2 – 0.3 cm

Table 1. Effects of salinity on survival, growth, total leaves grown, and total leaves shed (± SE) of 6-month-old *Rhizophora apiculata* seedlings in nursery

Salinity (‰)	0	5	10	15	20	25	30
Survival rate (%)	80.0±1.3	93.0±1.1	93.0±1.9	91.0±1.5	80.0±1.1	80.0±1.0	75.0±0.9
H (cm)	23.0^{a}±2.1	24.6^{b}±2.3	26.7^{c}±2.5	23.1^{a}±2.2	21.3^{d}±2.0	17.1^{e}±1.8	14.6^{f}±1.4
D_o (cm)	0.7±0.04	0.7±0.04	0.7±0.04	0.6 ±0.03	0.6±0.03	0.6±0.03	0.6 ±0.04
Total leaves grown	6.8^{a}±0.9	7.8^{b}±0.8	8.3^{c}±1.1	8.3^{c}±1.0	7.2^{d}±0.8	6.4^{e}±0.7	6.0e±0.7
Total leaves shed	2.0±0.2	0.7±0.2	0.4±0.1	0.6±0.1	0.2±0.1	0.1±0.0	0.1±0.0

H is stems height, D_o is stump diameter. Different letters (^{a,b,c}) in a line indicate difference of mean by univariate analysis of variance (ANOVA) and post-hoc test comparison at P = 0.05

Table 2. Effects of shoot depth fixed to the soil on survival, growth, total leaves grown, and total leaves shed (±SE) of 6-month-old *Rhizophora apiculata* seedlings in nursery

Depth of fruit shoot (%) fixed to the soil at sowing	50	33	25	0
Survival rate (%)	93.3±2.1	93.3±1.9	97.8±1.2	86.7±1.9
H (cm)	27.6^{a}±1.5	24.7^{b}±1.2	23.2^{c} ±1.0	15.5^{d}±0.8
D_o (cm)	0.73±0.05	0.74±0.04	0.73±0.03	0.62±0.02
Total leaves grown	13.2^{ab}±1.1	14.3^{b}±1.2	12^{c}±1.0	7.9^{d}±0.8
Total leaves shed	4.4^{a}±0.5	1.2^{b}±0.3	1.2^{b}±0.3	0.3^{c}±0.03

H is stems height, D_o is stump diameter. Different letters (^{a,b,c}) in a line indicate difference of mean by univariate analysis of variance (ANOVA) and post-hoc test comparison at P = 0.05

4.2 Plantation Establishment

Low tide (LT) area seems to be better than high tide (HT) area for survival rate and growth of *R. apiculata*. This may result from; firstly, daily flooding time to planted trees at the first six months in HT (0.5 – 3 hours/day) is longer than that in LT (0.3 – 1.5 hours/day); secondly, mudflat is more stable in LT compared to HT; thirdly, strength of sea wave is stronger in HT compared to LT [18]. Primary result in the present study indicated the potentiality of *R. apiculata* in invading new mudflats as indicated by Duke [20]. Contrarily, Lewis [3] indicated the unsuccessful planting mangrove forest trees in new mudflats of some other mangrove species. This indicates that selecting suitable species to new mudflats is the first priority. Selecting many species, which focused on creating mixed-mangrove forests toward ecological functions, may be not a good choice for new mudflats [3].

5. CONCLUSION

Mangrove forests have been establishing for shoreline protection, aquaculture development, and environment protection. Having healthy seedlings is a prerequisite for successful forest establishment. Fixing 50% fruit shoot to soil at sowing and seedlings watered by 10‰ salinity are recommended techniques for producing seedlings of *R. apiculata.* However, 5-month-old seedlings must be sea-watered for at least one month before planting. *R. apiculata* can grow well in new mudflats. However, low tidal regime areas as tide happens 100 – 299 days/year with water depth of 0.2 – 0.3 m in 4 – 8 hours/day are more suitable for successful mangrove forest establishment, where 36-month-old planted trees achieved 187.7 cm tall, 4.2 cm stump diameter, and 41.8 prop-roots per tree. Selecting species, which are suitable to new mudflats, is important. Since not all mangrove tree species can be ready colonizers to new mudflats.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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