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DIVERSITY OF SEED GERMINATION IN *XYLOCARPUS* SPECIES

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ABSTRACT

Xylocarpus species of mangroves are present in coastal areas of the World including India. The *Xylocarpus granatum* is distributed in very few numbers in coastal areas of Krishna and Godavari regions. On the other hand *Xylocarpus moluccensis* is distributed in high number in the above regions. The mangrove forest distribution is effected by many factors, including excessive wood gathering, fish pond operations, diversion of fresh water and coastal development-ports, industries etc. As there is vast variation in the distribution of *Xylocarpus granatum* and *Xylocarpus moluccensis* in the above regions. We tried to understand the possible causes for narrow distribution of *Xylocarpus granatum*. As it is known that seed germination, dormancy and dispersal, and other environmental factors play a vital role in the distribution and establishment of these species. Basing on the above speculation, we performed experiments to find out whether there are any innate differences in seed biology of *Xylocarpus* species. We considered three important factors for seedling establishment of *Xylocarpus granatum* such as water levels of the tide, seed size and position of the seed in the soil. The results of *Xylocarpus granatum* showed that the low water level (3-2 cm) resulted in high root length (5.2 cm) when compared to other water levels and large size of seeds formed a large number of roots and radical side of the seed should be intact with soil for root initiation. Whereas, *Xylocarpus moluccensis* not shown this criteria. Our results indicated that there are differences in the response to water levels of tide, seed size and position of seed in the soil among two species of *Xylocarpus*. Differences in the germination pattern among two species also reflected in photosynthetic performance of plants. Above data indicates that innate genetic potential of *Xylocarpus moluccensis* may be contributing to their distributional success. It also indicates that root formation and elongation are crucial for establishment of seedling in *Xylocarpus granatum* species which could be a reason for its narrow distribution.

Key words: ***Xylocarpus granatum*, seed germination, root development, seedling establishment**

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INTRODUCTION

Xylocarpus granatum and *Xylocarpus moluccensis* belongs to the family Meliaceae. *Xylocarpus granatum* is a medium sized tree with well developed woody trunk and fruit, and the bark is thin and peels out into flanks or long patches. The trunk base is smoothly round with ribbon like serpentine roots, without pneumatophores. Whereas *Xylocarpus moluccensis* contains buttress roots with cone shaped pneumatophores. *Xylocarpus granatum* was rediscovered after 9 decades and categorized as critically endangered plant (IUCN) in Maharashtra and the west coast of India[1]. Its rarity and endangered nature in Malaysia and other states like Kerala and Tamilnadu has been reported [2,3].Field survey during 2000-2002 in east coast Godavari mangroves indicated that *Xylocarpus granatum* is rare and *Xylocarpus moluccensis* is distributed well[5]. Recent observations revealed that *Xylocarpus granatum* is completely absent in Godavari mangroves. Availability and distribution of *Xylocarpus granatum* in Krishna mangroves were reported in Kothapalem, Nizampatnam and Diviseema reserve forests[6,7]. The field survey indicated that there is a vast variation in the distribution of *Xylocarpus granatum* and *Xylocarpus moluccensis* in the Krishna and Godavari mangroves. In the present work, we tried to understand the possible causes for narrow distribution of *Xylocarpus granatum*. As it known that seed dormancy and germination, and other environmental factors play a vital role in the distribution and establishment of these species. Early seedling establishment is crucial for successful restoration of mangrove forest. Basing on the speculation, we performed experiments to find out whether there are any innate differences in the seed biology and response to water levels of *Xylocarpus* species We considered three important factors for seedling establishment of *Xylocarpus granatum* such as water levels of the tide, seed size, and position of the seed in the soil. When seedlings of both species were established in our botanical garden, many of the young seedlings survived in spite of entirely different environment. We wondered whether there are any rapid physiological adjustments or differences between these two species, which stems from their differences in seed germination pattern. We also wondered whether these are true mangroves or not, but literature indicate that they are true mangroves. It is of interest to investigate whether the observed differences in the seed germination pattern reflect in physiological performance of the plants. Physiological robustness may provide a means for conservation programme. Indeed, we did find out some innate differences between these two species.

MATERIALS AND METHODS

Collection of plant material

Xylocarpus granatum and *Xylocarpus moluccensis* fruits, seeds, stem cuttings and young seedlings were collected from Kothapalem and Aduvuladeevi regions of Krishna mangroves during period of September 2013.

Water treatment for seed germination

Seeds were sown in tap water at different water levels in 5 plastic trays filled with tap water in the range of 1-5 cm. Each tray contains 10 seeds and germination pattern was observed for 20 days, variations in seed germination, root length, root growth and survivability with respect to water levels were recorded.

Preparation of soil bags

Soil used for seed germination and seedling growth, contains red soil, sand and ash in 60:30:10 ratios respectively. Very young seedlings were collected from above mangrove areas and transplanted on to above soil bags and maintained them in Botanical garden of Acharya Nagarjuna University. We observed variations in seed germination pattern depending upon seed position in soil bags. We checked the seed positional effect on germination in the soil by keeping the seeds of both species by allowing the seed to be in radical side towards the soil in one case and opposite side of the radical towards the soil in another case and recorded observations.

Assessing the photosynthetic performance of both species

We assessed the photosynthetic performance of both plant species established in our botanical garden with the help of ADC bio scientific L pro-SD advanced photosynthetic measurement system (ADLBSL). Photosynthetic rate under various light quanta for both species were recorded under ambient CO_2 and relative humidity levels.

RESULTS

Proper seed germination and seedlings establishment are vital for seedling survival. This depends upon nature and number of roots. In our study, we observed the rate of seed germination in both species as 90% (Table 1), but the emergence of roots and rate of survivability is only 50% in *Xylocarpus granatum* and 100% in *Xylocarpus moluccensis* (Table 2). The seed size is playing a major role in seed germination as size of seed increased it produced more number of roots visa versa in the case of small sized seeds (Fig 1).

Table 1
Variation in germination between X. moluccensis and X. granatum

Plant Name	No .of seeds sown	No. of seeds germinated	Mean	SE±SD
X. moluccensis	20	18	87.5	17.01
X. granatum	20	18	81.2	20.25

Table 2
Variation in survivability between X. moluccensis and X. granatum

Plant Name	No. of seeds germinated	No. of seedlings survived	Mean	SE±SD
X.moluccensis	18	18	98.5	20.25
X.granatum	18	09	55.5	21.23

Figure 1
Effect of seed size on X. granatum germination and root initiation
(1 cm bar = 11.25 mm)

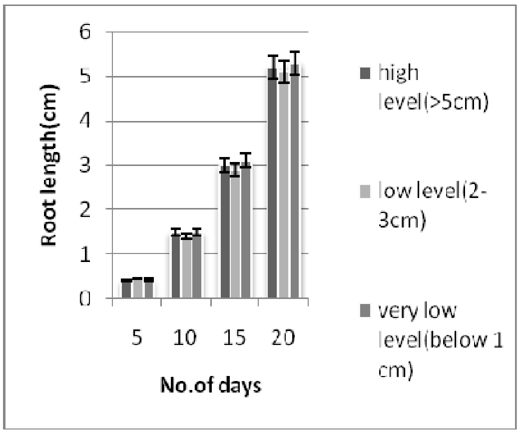

Figure 2
Effect of water height on seed germination in X. moluccensis

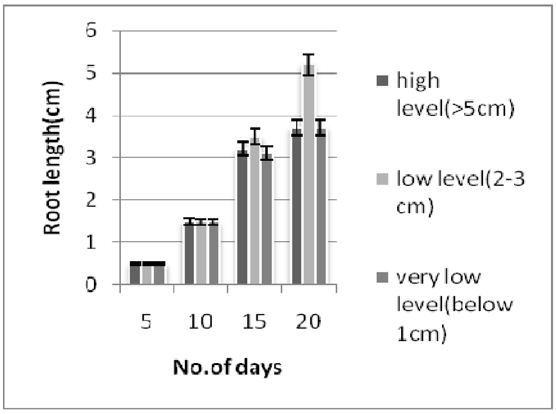

Figure 3
Effect of water height on seed germination in X.granatum

No variation in root length could be detected in the initial 5-10 days among high, low, very low water levels in *Xylocarpus granatum* plants (Fig.3). After 15 days, root length increased in low water level treated seeds of *Xylocarpus granatum* (Fig.3). No variation could be detected in root length among the seeds exposed to three levels of water in *Xylocarpus moluccensis* (Fig.2). And very low water level seedling roots wilted and some died. We observed that when the roots are exposed to external environment roots wilted and could not function again.

(a) **(b)** **(c)** **(d)**

Figure 4
a) X. moluccensis seed germination, b) White color roots of X.granatum
c) Roots exposed to environment, d) Blackening of roots
(1 cm bar = 20 mm).

Primary roots of *X. granatum* are very sensitive and delicate. When they exposed to external atmospheric conditions they turn into black color and could not function again (Fig.4b-d). This nature was not observed in *X. moluccensis* where no water is required for seed germination and for root growth (Fig.4a). Position of the seed also plays an important role in seed germination in *X. granatum* and the radical side of the seed should be intact with soil (Fig.5b) for successful germination (where roots are not exposed to air), whereas in case of *X. moluccensis* there is no particularity of seed position needed for good germination(Fig.5a).

(a) **(b)**

Figure 5
Radicle positions (a) X. moluccensis (1 cm bar = 6. 4 mm), (b) X.granatum (1 cm bar = 1.75 mm).

A **B** **C** **D**

Figure 6
A) Seedling of X. moluccensis (1 cm bar = 2.5 mm).
B, C& D seedlings of X. granatum (1 cm bar = 6.75 mm).

When *Xylocarpus granatum* seedlings were transferred from mangrove forest on to soil bags containing soil and sand in 70:30 ratio, the shoot tip turned into black color and become necrotic, activating the axillary buds and growth of lateral branches (Fig 7b). In case of *Xylocarpus moluccensis,* this kind of necrotic sensitivity could not be detected (Fig 7a).

a b

Figure 7

Lateral bud initiation (1 cm bar = 6.0 mm)

a b

Figure 8

Young seedlings growing in botanical garden of A.N.U.
a) X. moluccensis b) X.granatum

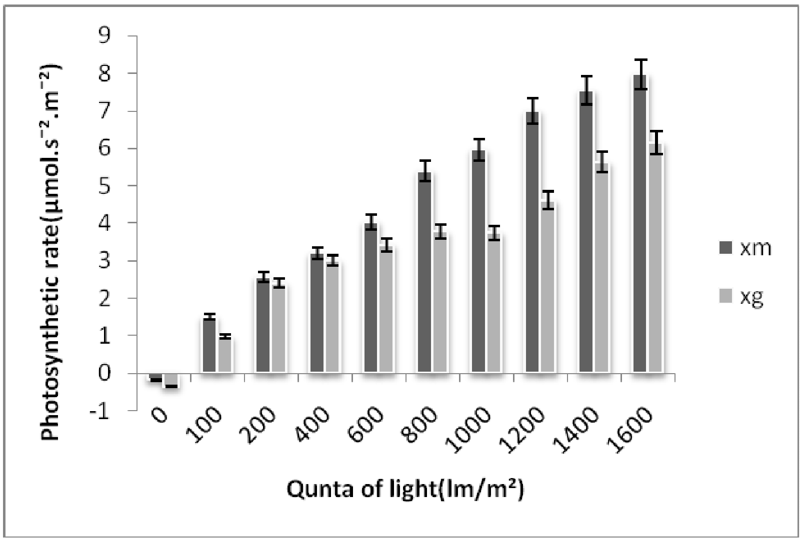

Figure 9

Variation of photosynthetic rate between X.granatum and X.moluccensis

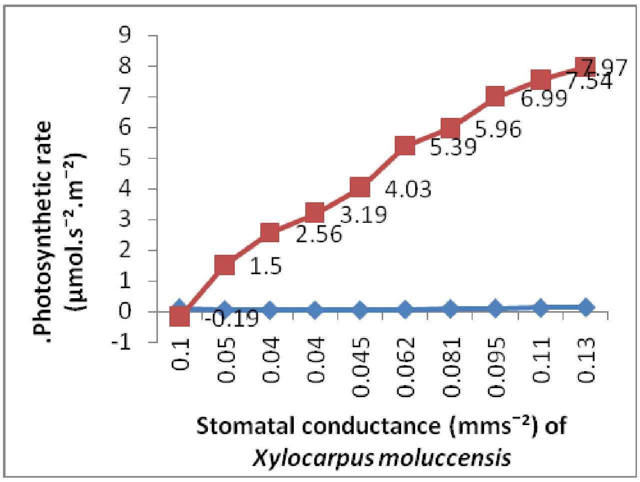

Figure 10

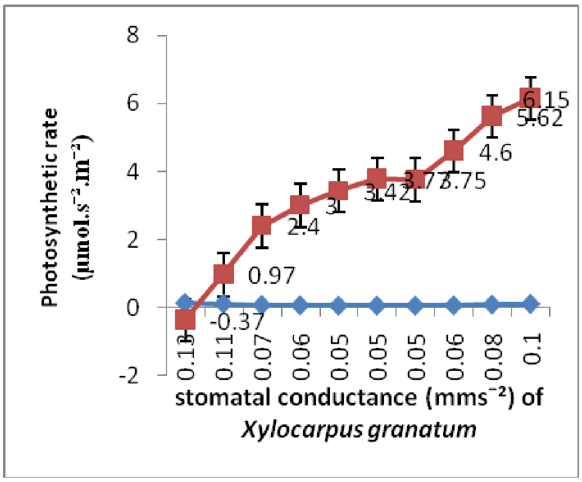

Figure 11

The above data indicates that X. granatum was found to be having less photosynthetic rate and stomatal conductance at all levels of light quanta, when compared to X. moluccensis (Fig 9-11).

DISCUSSION

Most of the mangrove plants are viviparous, few are non viviparous. Viviparous plant seeds can able to survive against the harsh mangrove habitat. Whereas non viviparous plant seeds are unable to germinate in their natural habitat due to different seed germination problems such as high salinity, predation, viability, light limit and shading. Seed germination and growth are effected by dehydration, flooding, salinity and tides[8, 9]. *Xylocarpus granatum* and *Xylocarpus moluccensis* are non viviparous plants. *Xylocarpus granatum* plants can survive against flooding, salinity and shade conditions [9]. Seeds are floating well [10]. Seed germination is widely reported to be affected by seed size, position of seed in the soil and different water levels on the soil. The seeds of *Xylocarpus granatum* showed clear preference for low water levels from 3 cm to 2 cm. And water level above 5cm significantly decreased root development and seedling establishment. High water levels could inhibit germination either by low oxygen availability or water contents [11]. Effect of high and very low water levels are serious to seedling establishment and water levels above 5cm was deadly to *Xylocarpus granatum* seeds under which no seedling succeeded to establish, when compared to *Xylocarpus moluccensis.* Likewise, root initiation of *X.moluccensis* was not evidently depressed by different water levels (Fig.4 a). We observed that at high water level (above 5 cm) roots will not grow to certain lengths (< 3.2 cm) as well as seeds are float on the water. Due to contractions of soil by fluctuated water levels, delicate roots may be damaged. Due to that, seedling establishment does not occur properly. Thus, seedlings are unable to survive at high flooding water. At very low water level (< 1 cm) seeds germinate as well as root length also high. But due to fluctuations of water levels, arising roots may be exposing to external environment, which will result in wilting and death of roots. Due to less number of roots, seedlings intolerant to flooding effect. A small increase in root length leads to a large increase in stability during the earliest seedling stage [12]. Seedling dislodgment was linearly related to the length of the longest root of seedling[12]. The anchoring of propagule will be more difficult, especially on the seaward side when wave action is highest and the necessity for propagule to entangle in certain root structure is very important [13]. At low water levels (3 cm to 2 cm) root system was developed well and root length also high (4.5 cm to 5.2 cm). Because of that, roots are penetrating into the soil deeply; as a result seedlings make it strong. This type of seedlings can survive against tidal effect, whereas the mangrove field has never maintains constant water levels, even though without water content, *Xylocarpus moluccensis* can able to germinate and establish greatly.Seed size also plays a critical role in seedling establishment. Propagule size is also linked with the rapid establishment through the formation of root to anchor in the substrate. Large propagule contains high amounts of reserves, which can provide energy during root formation[14.] We observed that mostly large number of roots arising from big sized seeds and small sized of seeds contains less root number. 90% of the seeds are established as seedlings which are having more number of roots and high root length (Fig.1). Position of the seed in soil also plays an important role in seed germination; *Dipteryx panamenis* with the radical end down is recommended [16]. Seedling establishment of *Xylocarpus* plants radical side of the seed always should be in contact with soil, not exposed to external environment. Whereas in the case of *X.moluccensis* it was not the case (Fig.5a). Frequent tidal effect of mangrove habitat may change the position of seeds. Due to this reason, *X.granatum* plants might have reduced in numbers when compared to *X.moluccensis*. The observed differences in seed germination patterns also reflected interns of differences in photosynthetic rate and stomatal conductance reflecting the genetic basis for the above differences. Hence, the observed the physiological and seed biology differences may be a reflection of evolutionary selection pressure on the respective genomes.

CONCLUSION

Xylocarpus granatum is highly sensitive to high water level and vary low water levels, especially at early development stage, the favorable water levels range for seed germination at 3 cm to 2 cm from the soil. In the position of seed, radical should be connecting with soil. Wilted nature of primary roots, position of seed in the soil may be a difficult situation for establishment of *X. granatum* when compared to *X. moluccensis*. The adverse water levels in the field might act as a primary obstacle for natural regeneration of the species. The size of the seed is determining factor for proper growth. Big sized seeds are favorable for proper seed germination and seedling establishment. Photosynthetic performance of plants reflected seed germination pattern. Thus, artificial breeding and culture should be adopted to ensure a higher survival rate of *X. granatum* seedlings. Due to these difficulties, seedling establishment at botanical garden of ANU took up to 3-4 months. The above studies are a step towards understanding the distributional and propagation difficulties of an important mangrove plants. We believe that understanding the bottlenecks for establishment and distribution problems will certainly pay a way for evolving effective conservation strategies.

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