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Short Communication

In vivo induction of multiple shoots for scaling up of propagation of tree mangrove *Bruguiera gymnorrhiza* (Linn.) Sav. (Rhizophoraceae)

*U. C. Basak & P. Das

Regional Plant Resource Centre, Bhubaneswar-751015, 0rissa, India

[*Email: u_c_basak@hotmail.com]

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In vivo shoot induction in hypocotyls and rooting of the regenerated shoots of *Bruguiera gymnorrhiza* (Linn.) Sav., was standardised as a new method of vegetative propagation. Rooting response of the air-layers made from multiple shoots with the aid of exogenously applied plant growth regulators (IAA+IBA) by pre-girdling is studied. Multiple shoots were induced in 80 % of the hypocotyls treated and the number of regenerated shoots varied from 2-5 with an average value of 3.5 per hypocotyl. Better rooting performance (80 %) was recorded in air-layers treated with IAA+IBA (3000 mg l^{-1} each). More than 80 % of the rooted air-layers survived in the nursery in polybags. Thus, through this method, 2-3 times more planting materials could be made available for vegetative propagation of *B. gymnorrhiza.*

[**Key words :** *Bruguiera*, multiple shoots, propagation]

Species of *Bruguiera* Sav. (Rhizophoraceae) of the Mahanadi delta of Orissa, east coast of India are threatened largely due to human interference (for fuel and timber), encroachment of mangrove forests (for cultivation and shrimp culture) and various other uses[1]. *Bruguiera* commonly grows along the creeks in the intertidal zone in the Mahanadi delta and Bhitarkanika along the east coast of India. Natural regeneration is restricted due to depletion of growing stock, post-dispersal predation of seeds by insects and crabs[2], sporadic flowering and poor seed set. Present study is on *in vivo* multiple shoot induction and rooting in the segmented shoot for propagation to augment the supply of planting materials for large scale plantations of *B. gymnorrhiza* (Linn.) Sav.

Mature and healthy hypocotyls of *B. gymnorrhiza* were collected from mangrove forests in the Mahanadi delta of Orissa (20° 4′-20° 8′ N and 86° 45′-86° 50′ E) during June, 2000. About 100 hypocotyls of nearly equal length and diameter were decapitated in the collar region and were planted in earthen pots (30 cm × 20 cm) containing a mixture of garden soil and sand (1:1) and then placed on the propagation bench under green house conditions (temperature 30 ± 4° C and relative humidity 80 ± 5 %). The propagules were fed everyday with fresh water. Observations were made on the development of multiple shoots from the decapitated end of the hypocotyls.

Ninety-day-old shoots (10-15 cm) with 4-6 nodes were air-layered following randomised block design with three replications having twenty shoots per replication. Pre-girdling was done by removing a ring of bark (about 2.0 cm wide) below the basal node of the regenerated shoot. The nodal region above the girdled zone was treated with IAA and IBA alone or in combinations at 3000 mg l^{-1} each in talc [1,3] and covered with moist sphagnum moss wrapped in white polythene film. There were 4 treatments including the control. The rooting percentage and mean root number per air-layer were recorded after 80 days of treatment i.e. the expected rooting period[1]. ANOVA[4] was performed on number of air-layers rooted and mean root number per air-layer in each treatment to analyse variations among treatments and replications.

The hypocotyls of *B. gymnorrhiza* developed multiple shoots after removal of the plumular apex; 80 % of the hypocotyls gave rise to a number of multiple shoots 3.5 ± 0.1 per hypocotyl varying from 2 to 5 shoots per hypocotyl (Fig. 1).

The treatment having the combination of IAA and IBA induced more roots in the pre-girdled air-layers than any auxin applied alone. The auxin conjugate showed 80 % rooting whereas treatment with IAA or IBA alone resulted in poor rooting (Table 1). The untreated air-layers (without auxin), however, failed to root. Significant variations were noted in rooting of

Table 1—Rooting performance of multiple shoots developed from hypocotyl of *Bruguiera gymnorrhiza*
[Values in the parenthesis indicate angular transformation]

Treatment	Mean rooting percentage		Mean root (no./air-layer)	Survival (%)
Control	0	(0)	0	0
IAA (3000 mg l^{-1})	10	(18.4)	1.4 ± 0.16	40
IBA (3000 mgl^{-1})	33.3	(35.23)	1.5 ± 0.10	65
IAA + IBA (3000 mg l^{-1} each)	80	(63.4)	1.8 ± 0.12	80

Fig. 1— (A) Multiples shoots developed from single hypocotyl of *Bruguiera gymnor*rhiza and rooting through air-layers, (B) sapling with fibrous root system favourable for plantation.

the air-layers due to different treatments ($p < 0.0001$). The root number per air-layer, however, did not vary significantly.

The decapitation method for induction of axillary shoot bud in mangroves was reported for the first time especially in the context of vegetative propagation. Adventitious root formation in other species of Rhizophoraceae i.e. *B. parviflora* was reported earlier by Basak *et al.*[1,5] with exogenous application of auxins. The advantages of pre-girdling on rooting of shoots of tree mangroves were also reported[1,3]. This method of multiple shoot regeneration with a two to three fold increase may be used for production of propagules in the Rhizophoraceae mangroves for aforestation programmes.

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