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RESEARCH ARTICLE

STANDARDIZATION OF PROPAGATION METHODS FOR SOME THREATENED MEDICINAL PLANTS OF NORTH EAST INDIA

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ABSTRACT

The present study was undertaken to standardize the methods of propagation for some threatened medicinal plants of North East India. Three endangered species of medicinal plants of North East India *viz. Smilax china*, *Plumbago indica* and *Rhynchostylis retusa* were selected for the study. The results indicated that Stump cutting of *Smilax china* and Shoot Tip cutting of *Plumbago indica* recorded the shortest days to root initiation in 19.67 and 12.33 days, shoot emergence in 18.33 and 7.38 days and to attain 30cm height in 54.50 and 129.00 days, respectively. The study also revealed that the highest number of roots per cutting after 6 months both in *Smilax china* and *Plumbago indica* were 27.17 and 13.67 numbers respectively. In *Rhynchostylis retusa*, Cocohusk block + Charcoal bed (under shade net) gave good results in producing number of leaves per plant and roots per plant after 8 month were 7.33 and 7 numbers respectively.

INTRODUCTION

Medicinal plants constitute a considerably large component of natural vegetation. They have a great demand not only in the domestic level but also in the commercial market, mostly in the herbal and pharmaceutical industries. A large number of medicinal plants species are considered threatened due to such high demand and destructive collection practices. Thus, the conservation efforts are needed to save these species in the wild by maintaining their wild populations. Propagation of such threatened species in the nurseries is a means towards conserving their genetic stocks. Propagation of medicinal species is of great importance, because as cultivation is increased, extraction is discouraged, thereby promoting the sustainable management of these species (Maroni *et al.,* 2006). Through the nurseries it is possible to make available planting material of these species for commercial cultivation and use in herbal industries. However, in the absence of proper information on the propagation techniques, nursery management and cultivation technology of medicinal plants, such initiatives to establish medicinal plants nurseries and cultivate them as commercial crops will not be successful.

The genus *Smilax* occurs in tropical and temperate zones in all the continents. They are dioecious monocotyledons with petiolated leaves, reticulated nervation, growing as climbing herbs or scandent sub-shrubs (Andreata, 1980). The leaves are elliptically oblong to subrounded, 5 to 8 centimeters long, 2.5 to 4 centimeters wide; those toward the end of the branches are much smaller, and are veined. The petioles are about 7 millimeters long, with adnate apiculate stipules, which frequently are extended into tendrils. The inflorescence arises from the upper leaf axile, 3 to 5 centimeters long. The flowers are yellowish-green, their pedicels subtended by bracteoles, umbellate. The berries are globose, and reddish when ripe. The dried rhizome of *Smilax china* L. of the family *Smilacaceae*, known as *Chobchini* in Hindi, contains fat, sugar, glycoside, coloring matter, saponin, gum, tannin, cinchonin, smilacin and starch (Nadkarnis, 2002) and it exhibits antiinflammatory, diuretic, anti-diabetic, anti-psoriatic and digestive properties (Shao *et al.,* 1992). In the past, in folk medicine, plants of the genus *Smilax* had been used extensively worldwide for the treatment of many diseases (Schroeder, 1988). The interest in the genus has increased again after the discovery that the Smilacaceae family produces large amounts of steroidal saponins (Bernardo *et al.,* 1996).There are no studies on rooting of cuttings in *Smilax* species, but regarding seed germination there are few species studied and the tested variables are very distinct making it difficult to establish a comparative analysis of responses (Andreata and Pereira, 1990).

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Plumbago indica is a multipurpose medicinal herb of family Plumbaginaceae. A native of South Asia, the species is distributed throughout most of the tropics and subtropics; growing in deciduous woodland, savannas' and scrublands from sea level up to 2000 m altitude (Aditi *et al.,* 1999; Lubaina *et al.,* 2011 and Vishnukanta *et al.,* 2010). Its leaves are simple, alternate, oblong, short-cuneate at the base passing into a very short amplexicaul, exauriculate, reddish petiole; flowers pinkish to bright red, in long terminal spikes. Root contains Plumbagin (5-hydroxy-2-methylnaphthalene- 1,4-dione), sitosterol glycoside. Plumbagin is present in all the varieties of *Plumbago* to a maximum of about 0.91%. In India it is usually used to treat fever or malaria. Pharmacological studies have indicated that *P. zeylanica* extract has antiplasmodial (Simonsen *et al.,* 2001), antimicrobial (Ahmad *et al.,* 2000), antifungal (Mehmood *et al.,* 1999), anti-inflammatory (Oyedapo, 1996), hypolipidaemic and antiatherosclerotic activities (Sharma *et al.,* 1991). In Assamese the species is called "*Agyachit*".

Rhynchostylis retusa (also called Foxtail Orchid) is a wild epiphytic orchid in Assam and culturally very intrinsically related to the Assamese people. The orchid has a bunch consisting of 100 more than pink-spotted white flowers. The plant is found in semi-deciduous and deciduous dry lowland forests woodlands at elevations of sea level to 700 m, native to Malaya, Singapore, Thailand, Sri Lanka and India. They have stout, repent, short stem carrying to 12, curved, fleshy, deeply channeled, keeld, retuse apically leaves and blooms on an axillary, pendant, to 60 cm long, racemose, densely many flowered, cylindrical inflorescence that occurs in the winter and early spring. The plants die if the leaves are wet frequently. They prefer moderate waterings, heavy fertilizing, and very bright shade to direct morning light. Flowering is usually summer into fall. In Assam, it is popularly known as *Kopou Phool*, and the pink inflorescence is an integral part of a Bihu dancer's attire

MATERIALS AND METHODS

Experimental site

The experiments were under taken in the Medicinal and Aromatic Farm, Department of Horticulture, Assam Agriculture University, Jorhat during the year 2013-2014. The experimental site is located at 26°47' N latitude, 94°12' E longitude and at an altitude of 86.8 m above the mean sea level. The experiment was laid out in Randomized Block Design (RBD) comprising of five treatments and 4 replications for *Smilax china* & *Plumbago indica* and seven treatments and 3 replications for *Rhynchostylis retusa*.

The collection of these threatened medicinal plants were done from different parts of North East India. Treatments and treatment combination used in the experiments were:

For *Smilax china* & *Plumbago indica*

T_1: Stem cutting (15cm)
T_2: Leaf bud cutting
T_3: Cutting with two internodes
T_4: Shoot Tip cutting
T_5: Stump cutting

For *Rhynchostylis retusa*

T_1: Cocohusk block (open)
T_2: Cocohusk block (under shade net)
T_3: Cocohusk block + Cocopeat (open)
T_4: Cocohusk block + Cocopeat (under shade net)
T_5: Cocohusk block + Charcoal bed (open)
T_6: Cocohusk block + Charcoal bed (under shade net)
T_7: Control (in woody plant stem/branch)

Observations on days to root initiation, shoot emergence, to attain 30cm height and number of roots per cutting has been recorded for both *Smilax china* & *Plumbago indica* and for *Rhynchostylis retusa* number of leaves and roots per plant has been recorded at monthly interval upto 6 and 8 months, respectively.

Statistical Analysis: Collected data were statistically analyzed by using PASW statistics 18 computer based software.

RESULTS AND DISCUSSION

Different treatments were significant influence on root initiation, shoot emergence and to attain 30cm height in case of *Smilax china* (Table 1). The minimum days required for root initiation, shoot emergence and to attain 30cm height was recorded in stump cutting 19.67, 18.33 and 54.50 respectively. The number of roots was found to be highly significant due to different treatments. From the data obtained (Table 2) till 6 months, it was observed that Stump cutting produced more number of roots (13.67) compared to other treatments in short time.

Table 1 Effect of different treatment on root initiation, shoot emergence and to attain 30cm height in *Smilax china*.

Treatments	Root initiation (Days)	Shoot emergence (Days)	To attain 30cm height (Days)
Stem cutting (15cm)	21.67	21.08	176.00
Leaf bud cutting	25.58	20.58	166.50
Cutting with two internodes	23.00	20.91	164.50
Shoot Tip cutting	21.42	21.67	156.50
Stump cutting	19.67	18.33	54.50
CD (5%)	**1.08**	**0.73**	**12.66**

Table 2 Effect of different treatmnt in *Smilax china* on Number of roots formation.

Treatments	Number of roots After					
	1 month	2 month	3 month	4 month	5 month	6 month
Stem cutting (15cm)	1.33	3.83	5.42	5.75	6.00	6.17
Leaf bud cutting	0.84	4.25	6.92	7.21	7.41	7.71
Cutting with two internodes	1.08	5.54	7.58	7.67	7.92	8.00
Shoot Tip cutting	4.17	10.50	12.25	12.58	12.92	13.08
Stump cutting	2.58	8.60	12.50	13.25	13.54	13.67
CD (5%)	**0.74**	**1.14**	**0.92**	**0.92**	**0.96**	**0.98**

Data pertaining to the days required for root initiation, shoot emergence and to attain 30cm height was significantly influenced by different treatments in case of *Plumbago indica* (Table 3). The minimum days required for root initiation, shoot

emergence and to attain 30cm height was recorded in shoot tip cutting 12.33, 7.38 and 129.00, respectively. The number of roots was found to be highly significant due to different treatments. From the data obtained (Table 4) till 6 months it was observed that shoot tip cutting produced more number of roots (27.17) compared to other treatments.

Table 3 Effect of different treatment in *Plumbago indica* on root initiation, shoot emergence and to attain 30cm height.

Treatments	Root initiation (Days)	Shoot emergence (Days)	To attain 30cm height (Days)
Stem cutting (15cm)	20.39	12.05	178.00
Leaf bud cutting	13.79	7.63	147.25
Cutting with two internodes	20.22	11.33	142.75
Shoot Tip cutting	12.33	7.38	129.00
Stump cutting	18.72	9.04	145.00
CD (5%)	**1.60**	**1.17**	**4.13**

Table 4 Effect of different Treatment in *Plumbago indica* on Number of roots formation.

Treatments	Number of roots After					
	1 month	2 month	3 month	4 month	5 month	6 month
Stem cutting (15cm)	5.17	8.17	10.58	17.25	19.92	21.38
Leaf bud cutting	7.80	9.17	12.88	20.33	21.67	23.33
Cutting with two internodes	7.29	9.33	12.83	18.17	21.25	22.89
Shoot Tip cutting	9.83	12.33	17.50	21.09	23.92	27.17
Stump cutting	8.08	10.08	15.75	18.79	20.59	22.54
CD (5%)	**1.03**	**0.90**	**1.31**	**1.24**	**6.86**	**1.94**

The results indicated that the number of leaves and number of roots per plant after 8 months was significantly influenced by different treatments in *Rhynchostylis retusa* (Table 5). The maximum number of leaves and roots was recorded in Cocohusk block + Charcoal bed (under shade net) 7.33 and 7.00, respectively.

Table.5 Effect of different Treatment on number of leaves and roots in *Rhynchostylis retusa* plant after 8 months.

Treatments	No. of leaves per plant	No. of roots per plant
Cocohusk block (open)	4.00	3.67
Cocohusk block (under shade net)	5.00	5.00
Cocohusk block + Cocopeat (open)	4.33	4.67
Cocohusk block + Cocopeat (under shade net)	6.00	6.67
Cocohusk block + Charcoal bed (open)	5.00	5.33
Cocohusk block + Charcoal bed (under shade net)	7.33	7.00
Control (in woody plant stem/branch)	4.33	5.00
CD (5%)	**1.26**	**1.20**

CONCLUSION

From the present study it can be concluded that Stump cutting of *Smilax china* and Shoot Tip cutting of *Plumbago indica* required shortest days to root initiation, shoot emergence and to attain 30cm height. The study also revealed that the highest number of roots per cutting after 6 months both in *Smilax china* and *Plumbago indica* can be obtained. In *Rhynchostylis retusa*, Cocohusk block + Charcoal bed (under shade net) gave good results in producing number of leaves per plant and roots per plant after 8 months.

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