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RESEARCH ARTICLE

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Clonal propagation technique for mass multiplication of Noni (*Morinda citrifolia*) seedlings

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ABSTRACT : A study was under taken to standardize the type of cutting, concentration of growth hormone and suitable rooting media to produce elite seedlings in *Morinda citrifolia* at Forest College and Research Institute, Mettupalayam. Investigation was carried out to standardize the treatments to induce root and shoot growth in different types of cuttings of *Morinda citrifolia* L. using two growth regulators (IAA, IBA) used in different concentrations (1000, 1500, 2000 and 2500 ppm) in two kinds growth medium (Sand and Coir) for different type of cuttings, 2500 ppm IBA treatment recorded its superiority in terms of maximum sprouting (85%), rooting (79%) and survival (70%) percentage, number of roots (15.83), root length (8.51 cm), number of shoots (4.00) and shoot length (9.43 cm) in leafy shoot cuttings, irrespective of rooting media used. From this current study it is concluded that, for macro clonal propagation of noni (*Morinda citrifolia* L.), either leafy shoot cuttings or hard wood cuttings could be treated with 2500 ppm IBA or semi hard wood cuttings could be treated with 1500 ppm IAA and could be planted in coir pith medium for the production of elite planting materials.

KEY WORDS : Noni, Macro clonal, Cuttings, Growth regulators, Growth medium

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INTRODUCTION

Noni, *Morinda citrifolia* L. (Rubiaceae), is a small, fruit-bearing, evergreen shrub or tree that now grows throughout the tropics. It is a traditional source of medicine, dye and food for indigenous peoples and it recently has been marketed internationally as a dietary supplement. *M. citrifolia* has the ability to regenerate from shoots or root suckers rather than seed producing small thickets or groves. Noni is relatively easy to propagate from seeds, stem or root cuttings (Hartmann *et al.*, 2002). *M. citrifolia* is gaining popularity among the farmers and local communities due to its market value and tolerant to salinity and brackish water (Singh *et al.*, 2005b).

However, Noni seeds have a problem of seed dormancy thus, limiting its commercial propagation through

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seeds, since the untreated seeds need several months to a year for germination (Ponnaiyan and Vezhavendan, 2005). Many research works were carried out only on medicinal uses of noni fruits and plants by several authors (Mathivanan *et al.*, 2006 and Singh *et al.*, 2005b). However, the scientific information available on seedling production and improvement of macro clonal propagation techniques are scanty. Hence, with this back ground, the present study was under taken for clonal propagation in Noni for mass multiplication.

EXPERIMENTAL METHODS

A study was under taken to standardize the type of cutting, concentration of growth hormone and suitable rooting media to produce elite seedlings in *Morinda citrifolia* adopting the following experimental methodology. Three types of cuttings namely leafy shoot cutting, semi hard wood cuttings, hard wood cuttings were taken from the noni plant grown at the Department of Medicinal and Aromatic crops, TNAU, Coimbatore.

The cuttings were further trimmed to a length of approximately 15 cm with five nodes. In case of leafy shoot cuttings, two leaves near the apical point were retained. All the leaves of semi hard wood and hard wood cuttings were removed. A slanting cut was given at the basal part of all the cuttings. Then the cuttings were first treated with 0.2 percent Bavistin and subjected to following root inducing hormone treatments by quick dip method.

Treatments:

T_0 - Control (No treatment), T_1 - Treated with 1000 ppm IAA, T_2 - Treated with 1500 ppm IAA, T_3 - Treated with 2000 ppm IAA, T_4 - Treated with 2500 ppm IAA, T_5 - Treated with 1000 ppm IBA, T_6 - Treated with 1500 ppm IBA, T_7 - Treated with 2000 ppm IBA and T_8 - Treated with 2500 ppm IBA.

The stock solutions of IAA and IBA were prepared by dissolving the weighed quantity of each hormone (*i.e.* 1.0, 1.5, 2.0 and 2.5 g/l, respectively) first in sodium hydroxide solution and made into known volume with distilled water. Then the cuttings were treated in the root inducing hormones by quick dip method.

After treating the cuttings, five cuttings in each replication treatment wise were planted in the following rooting media in portrays.

Rooting media:

M_1- Coirpith, M_2- Sand

Design : FCRD

Replications : 3

The cuttings after planting in rooting media were placed in mist chamber maintained 40 ± 2°C temperature and 80 % RH and the following observations were recorded at 90 days after planting.

Sprouting percentage:

The number of cuttings sprouted at the end of test period (90 days) were counted in each treatment replication wise and the sprouting percentage of the cuttings were calculated by using the following formula :

$$\text{Sprouting percentage} = \frac{\text{Number of cuttings sprouted}}{\text{Number of cuttings planted}} \times 100$$

Rooting percentage:

The number of cuttings rooted at the end of test period (90 days) were counted in each treatment replication wise and the rooting percentage of the cuttings were calculated by using the following formula (Gurumurthi and Bhandari, 1988).

$$\text{Rooting percentage} = \frac{\text{Number of cuttings rooted}}{\text{Number of cuttings planted}} \times 100$$

Survival percentage:

The number of cuttings produced root and shoot and survived at the end of test period were counted survival percentage of the cuttings were calculated by using the following formula :

$$\text{Survival percentage} = \frac{\text{Number of cuttings survived}}{\text{Number of cuttings planted}} \times 100$$

Number of roots per cutting:

The rooted cuttings in each treatment, replication wise were washed in water to remove the medium without disturbing the root system. Then the number of roots were counted and the mean values were expressed in numbers.

Root length:

The rooted cuttings in each treatment, replication wise were washed in water to remove the medium without disturbing the root system. The root length was measured from the point of initiation to the tip of roots and the mean values were expressed in centimeter.

Number of shoots per cutting:

The sprouted cuttings in each treatment, replication wise were taken and the number of shoots produced in each cuttings were counted and the mean values were expressed in numbers.

Shoot length:

The sprouted cuttings in each treatment, replication wise were taken and the shoot length was measured from the point of initiation to the tip of the shoot and the mean values were expressed in centimeter.

EXPERIMENTAL RESULTS AND ANALYSIS

Between two growth regulators (IAA, IBA) used in different concentrations (1000, 1500, 2000 and 2500 ppm) irrespective of rooting media leafy shoot cuttings treated with 2500 ppm IBA treatment recorded its superiority in terms of maximum sprouting (85%), rooting (79%) and survival (70%) (Table 1, 2 and 3), number of roots (15.83), root length (8.51 cm), number of shoots (4.00) and shoot length (9.43 cm) followed by hardwood cuttings for all the above mention characters (Fig. 1,2,3 and 4). The study carried out by Tiwari *et al*. (2016) in *Dilleniapentagyna* were showed the IBA (500 ppm) was the best treatment in root induction.

In contrast, semi hardwood cuttings perform better at lower concentration of IAA (1500 ppm) than other two concentrations of IAA and IBA at all concentrations for all the observed parameters. Similar findings were reported in *Dilleniapentagyna* by Tiwari *et al*. (2016) in which semi hard wood cuttings were performed well compared to hardwood cuttings.

The study revealed that the dose and type of hormones was an important factor in the rooting of cuttings as suggested by Nautiyal *et al.* (1991). Aslam *et al*. (2007) reported that IBA treatment at 2500 ppm performed superior in terms of callusing percentage, rooting percentage, number of roots and root length in *Taxusbaccata*.

Propagation of tree species through stem cuttings is a function of several factors one of which is medium of propagation (Nanda, 1970 and Harsh and Mathana, 1985). In the present study, two rooting media *viz.*, coir pith and sand were evaluated. Between media, all three cuttings planted in coir pith produced significantly higher percentage of sprouting, rooting, survival and root and shoot numbers and their length than sand media. Among three cutting planted in coir pith medium leafy shoot cuttings resulted in pronounced increased performance than other two cuttings.

In semi hard wood cuttings, coir pith medium recorded significantly higher values for sprouting percentage (57%), rooting percentage (49%), survival

Table 1: Effect of growth regulator and rooting media on sprouting on different propagating materials in Noni (90 DAP)

Rooting media (M)/ treatments (T)	Sprouting percentage								
	Hard wood			Semi hard wood			Leafy shoot		
	M_1	M_2	Mean	M_1	M_2	Mean	M_1	M_2	Mean
T_0	40 (39.15)	33 (35.01)	37 (37.07)	40 (39.14)	37 (37.22)	38 (38.18)	47 (43.07)	43 (41.15)	45 (42.12)
T_1	67 (52.77)	57 (48.85)	62 (50.81)	67 (54.99)	57 (48.84)	62 (51.91)	60 (50.85)	50 (45.00)	55 (47.92)
T_2	50 (45.00)	47 (43.07)	48 (43.96)	78 (62.32)	76 (61.04)	77 (61.18)	67 (54.99)	63 (52.77)	65 (53.88)
T_3	63 (53.07)	47 (43.07)	55 (48.07)	60 (50.77)	57 (48.84)	58 (48.84)	60 (50.85)	53 (46.92)	57 (48.88)
T_4	67 (54.98)	47 (43.07)	57 (49.03)	50 (45.00)	47 (43.07)	48 (43.07)	70 (56.99)	53 (46.92)	62 (51.96)
T_5	47 (42.70)	43 (41.15)	45 (41.93)	53 (46.92)	50 (45.00)	51 (45.96)	57 (48.84)	53 (46.92)	55 (47.88)
T_6	50 (45.00)	47 (43.07)	49 (45.51)	67 (55.07)	63 (53.06)	65 (54.07)	50 (45.00)	47 (43.07)	48 (44.03)
T_7	49 (44.67)	46 (42.58)	48 (44.03)	50 (45.00)	47 (43.08)	48 (44.03)	57 (48.84)	53 (46.92)	55 (47.88)
T_8	83 (66.14)	77 (61.22)	80 (63.68)	50 (45.00)	47 (43.08)	48 (43.08)	87 (72.19)	83 (66.14)	85 (69.17)
Mean	57 (49.03)	49 (45.51)	53 (46.86)	57 (49.03)	53 (46.92)	54 (47.43)	62 (52.13)	50 (45.00)	56 (48.56)
	S.E.±	C.D. (P=0.05)		S.E.±	C.D. (P=0.05)		S.E.±	C.D. (P=0.05)	
T	3.919	7.951*		3.348	6.791**		3.316	6.726**	
M	1.847	3.414*		1.178	2.010*		1.443	2.179*	
T x M	5.543	11.244**		4.735	9.604*		4.689	9.512**	

(Figures in parentheses indicate arcsine values) * and ** indicate significance of values at P=0.05 and 0.01, respectively

percentage (48%), number of roots (17.51), root length (8.63 cm), number of shoots (2.99) and shoot length (7.48 cm) then the sand medium, irrespective of auxin concentrations. The hard wood cuttings also recorded significantly higher sprouting percentage (57%), rooting percentage (40%), survival percentage (38%), number of roots (18.84), root length (9.44 cm), number of shoots (3.11) and shoot length (8.68 cm) in coir pith medium than the sand medium, irrespective of auxin concentrations. The same medium was also reported to be superior in *Sterculiaurens* by Reddy and Venkiah (1994).

Table 2: Effect of growth regulator and rooting media on rooting per cent of different propagating materials in Noni (90 DAP)

Rooting media (M)/ treatments (T)	Rooting percentage								
	Hard wood			Semi hard wood			Leafy shoot		
	M_1	M_2	Mean	M_1	M_2	Mean	M_1	M_2	Mean
T_0	30 (33.00)	23 (28.77)	27 (31.89)	30 (33.21)	27 (30.78)	29 (32.78)	40 (39.23)	37 (37.22)	39 (38.27)
T_1	40 (39.15)	37 (37.22)	38 (38.18)	47 (43.07)	43 (41.15)	45 (42.11)	47 (43.07)	42 (41.15)	45 (42.11)
T_2	43 (41.07)	27 (30.78)	35 (35.92)	73 (60.04)	67 (54.78)	70 (56.90)	53 (46.92)	50 (45.00)	52 (45.96)
T_3	47 (43.07)	27 (33.00)	37 (38.03)	47 (43.07)	33 (37.22)	42 (40.15)	57 (48.84)	43 (41.15)	50 (45.00)
T_4	37 (37.22)	27 (30.78)	32 (34.00)	43 (41.15)	37 (37.14)	40 (39.14)	57 (48.84)	53 (46.92)	55 (47.88)
T_5	30 (33.00)	24 (28.77)	28 (30.88)	47 (43.07)	33 (35.21)	40 (39.14)	47 (43.07)	43 (41.15)	45 (42.11)
T_6	30 (33.00)	27 (30.78)	29 (31.89)	67 (54.98)	60 (50.85)	63 (52.91)	50 (45.00)	47 (43.07)	48 (44.03)
T_7	33 (35.21)	30 (33.00)	32 (34.10)	47 (43.07)	37 (37.22)	42 (40.15)	47 (43.07)	43 (41.15)	45 (42.11)
T_8	67 (54.78)	53 (46.92)	60 (50.85)	37 (37.22)	33 (35.21)	35 (36.22)	80 (63.44)	77 (61.22)	79 (62.58)
Mean	40 (39.15)	31 (33.54)	35 (35.85)	49 (43.96)	41 (41.15)	45 (42.38)	53 (46.92)	48 (43.23)	51 (44.97)
	S.E.±	C.D. (P=0.05)		S.E.±	C.D. (P=0.05)		S.E.±	C.D. (P=0.05)	
T	3.286	6.667*		3.028	6.141**		2.212	4.487**	
M	1.549	3.142		1.427	2.705*		1.043	2.098*	
T x M	4.648	9.428*		4.282	8.685**		3.129	6.346**	

(Figures in parentheses indicate arcsine values) * and ** indicate significance of values at P=0.05 and 0.01, respectively

Table 3: Effect of growth regulator and rooting media on survival per cent of different propagating materials in Noni (90 DAP)

Rooting media (M)/ treatments (T)	Survival percentage								
	Hard wood			Semi hard wood			Leafy shoot		
	M_1	M_2	Mean	M_1	M_2	Mean	M_1	M_2	Mean
T_0	27 (30.78)	23 (26.68)	25 (28.77)	30 (30.78)	27 (30.78)	29 (30.78)	40 (39.23)	37 (37.23)	39 (38.27)
T_1	40 (39.15)	37 (37.22)	38 (38.19)	47 (43.08)	43 (41.15)	45 (42.11)	47 (43.08)	43 (41.15)	45 (42.12)
T_2	40 (39.15)	27 (30.78)	33 (34.97)	73 (59.00)	67 (54.78)	70 (56.89)	53 (46.92)	50 (45.00)	52 (45.96)
T_3	47 (43.07)	30 (33.00)	38 (38.03)	47 (43.07)	37 (37.22)	42 (40.15)	57 (48.84)	44 (41.15)	50 (45.00)
T_4	33 (35.21)	27 (30.78)	30 (33.00)	44 (41.15)	36 (36.83)	40 (39.14)	57 (48.84)	53 (46.92)	55 (47.88)
T_5	30 (33.00)	23 (28.77)	27 (30.89)	43 (41.07)	30 (33.00)	37 (37.03)	47 (43.08)	43 (41.16)	45 (42.12)
T_6	30 (33.00)	27 (30.78)	28 (31.89)	60 (50.85)	57 (49.22)	58 (50.03)	50 (45.00)	47 (43.08)	48 (44.03)
T_7	33 (35.21)	30 (33.00)	32 (34.11)	47 (43.07)	37 (37.22)	42 (40.15)	47 (43.07)	43 (41.16)	45 (42.11)
T_8	67 (54.78)	53 (46.92)	60 (50.85)	37 (37.22)	30 (33.00)	33 (35.11)	73 (59.01)	67 (54.78)	70 (56.89)
Mean	38 (37.92)	31 (33.33)	35 (35.63)	48 (43.07)	41 (40.79)	44 (41.93)	52 (45.86)	47 (43.08)	50 (45.00)
	S.E.±	C.D. (P=0.05)		S.E.±	C.D. (P=0.05)		S.E.±	C.D. (P=0.05)	
T	3.157	6.405*		3.581	7.265*		2.201	4.465*	
M	1.388	2.819*		1.688	2.821*		1.037	2.048*	
T x M	4.465	9.058**		5.065	10.274**		3.113	6.315**	

(Figures in parentheses indicate arcsine values) * and ** indicate significance of values at P=0.05 and 0.01, respectively

The role of rooting media in inducing rooting and developing healthy seedlings in stem cutting is well known. An ideal rooting media should be able to provide enough moisture and nutrients for root initiation and further development besides preventing the desiccation of the cut ends. Similar findings on performance of coir pith media was reported by Nanda (1970) for propagation of *Casuarinaequisetifolia*, *Azadirachta indica* and *Ceibapentandra*.

However, as per Casimiro *et al*. (2003) studies the inclusion of IBA in the medium increased the rooting percentage, number of roots per rooted shoot and length of the roots. Many classic and recent studies have shown thatauxin plays a central role during lateral root

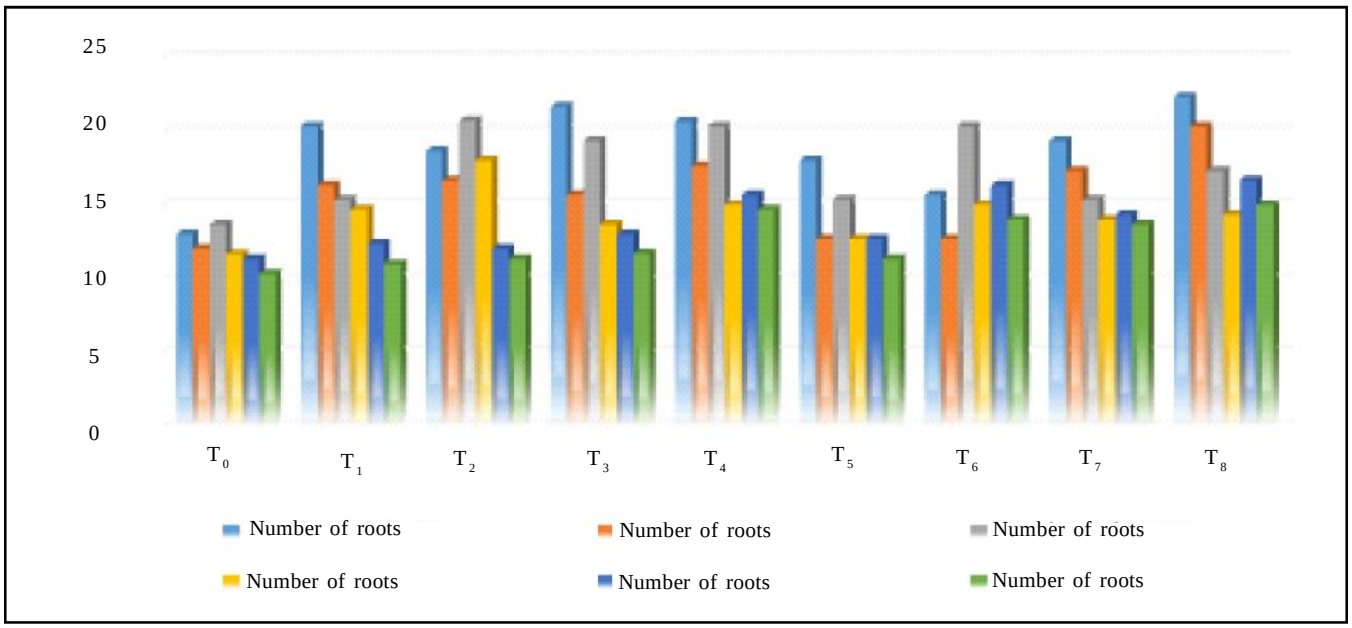

Fig. 1: Effect of growth regulator and rooting media on number of roots of different propagating materials in Noni (90 DAP)

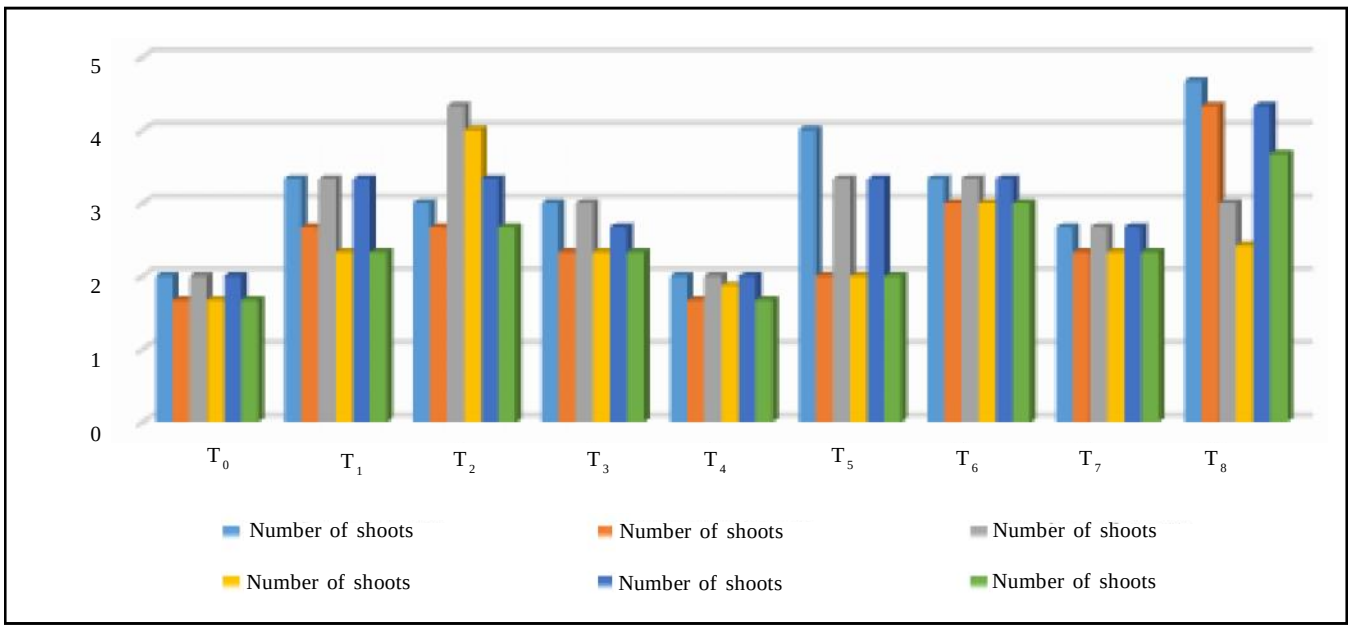

Fig. 2: Effect of growth regulator and rooting media on number of shoots of different propagating materials in Noni (90 DAP)

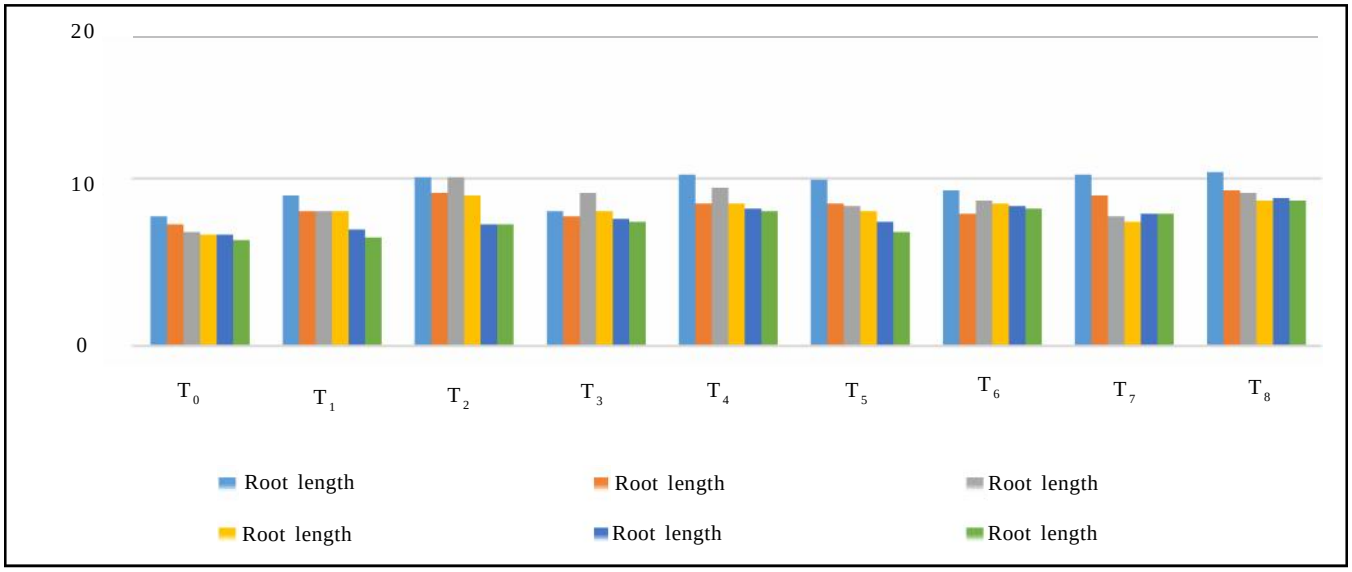

Fig. 3: Effect of growth regulator and rooting media on root length of different propagating materials in Noni (90 DAP)

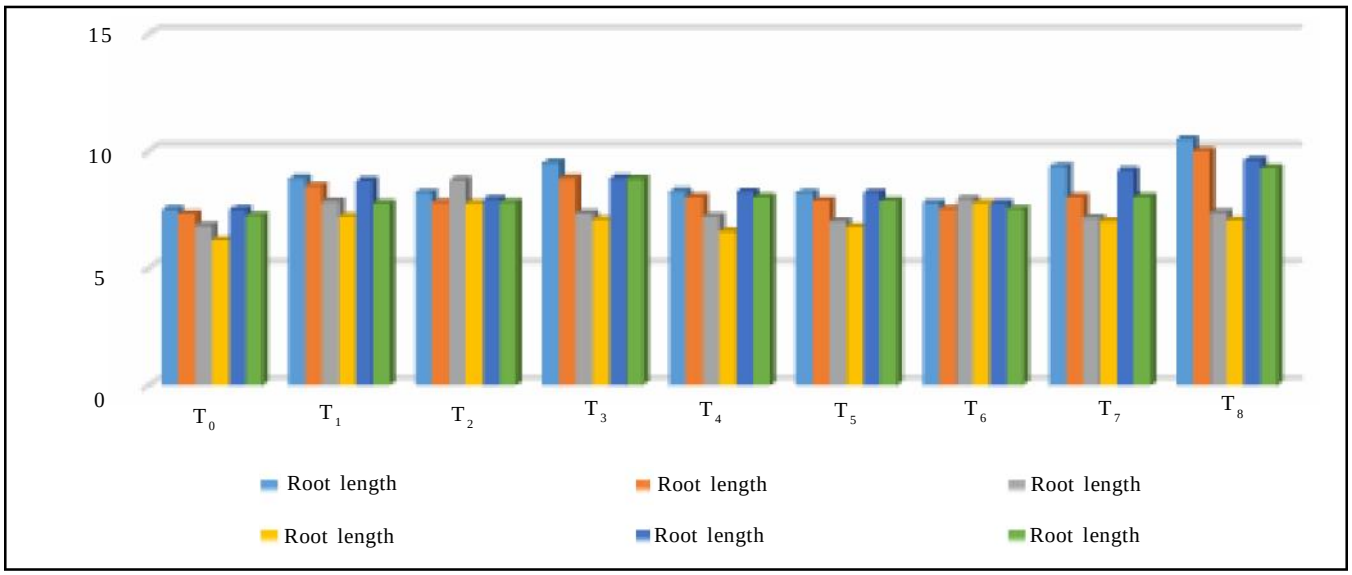

Fig. 4: Effect of growth regulator and rooting media on shoot length of different propagating materials in Noni (90 DAP)

development.

Conclusion:

In a holistic perspective, the current study revealed that, for macro clonal propagation of *Morinda citrifolia,* either leafy shoot cuttings or hard wood cuttings could be treated with 2500 ppm IBA or semi hard wood cuttings could be treated with 1500 ppm IAA and planted in coir pith medium for the production of elite planting materials.

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