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RESEARCH ARTICLE

ROOTING OF ROOT CUTTINGS: A BREAKTHROUGH IN VEGETATIVE PROPAGATION.

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Key words:-

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Abstract

Regeneration of plantlets from adventitious roots is a new idea, which is achieved through the present study using prop roots of *Clusia fluminensis*, a neotropical ornamental species. Three IBA (Indole 3-butyric acid) concentrations in ppm level were used for the present study in order to establish the effect of rooting and SVI (sprouting value index) was employed to interpret the efficiency of management practices. Field experiments were carried out using three rooting media for three consecutive years in a tropical environment. The results obtained indicate high SVI (sprouting value index), when prop root cuttings planted in root trainers with coir pith compost (RTCP) for all the three concentration of IBA applied. Sprouting percentage was increased and the delay in completion of sprouting/rooting initiation decreased by the use of IBA treatment. Increased percentage of (CWR) callus without rooting and (SUP) sound unsprouted propagules percentages indicates defects in management practices, dormancy and or the genotype of the cultivar. Even though the rooting of cutting technology using exogenous application of IBA is well established, raising plantlets by rooting of root cuttings will be new knowledge in artificial propagation studies.

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Introduction:-

Plant propagation has a global effect, as it is a fundamental occupation of human kind and its discovery dates back to the origin of civilization and ornamental gardening attained high level during the period between 500 B.C to A.D. 1000 (Harlan, 1992; Solbrig, 1994). The vegetative propagation of herbaceous plants can be performed easily as many of them produces bulbs, rhizomes, corms, tubers, offsets, suckers and stolons which are natural vegetative propagating structures. There are various reports on exogenous application of auxin on stem cuttings and its significant positive effect in adventitious rooting initiation in various tree species (Hartmann, *et al.*, 1997; Palanisamy, *et al.*, 1998; Hossain, *et al.*, 2004; Singh, *et al.*, 2004). It is obvious that the process of root and shoot regeneration from adventitious roots is a novel method, which is being achieved through the present investigation. *Clusia fluminensis* is an ornamental tree with entire, leathery and succulent leaves with obligate crassulacean acid metabolism (Lüttge, 1999) and remarkable ecophysiological plasticity (Lüttge, 2007). The lack of technology for regeneration from adventitious roots and ornamental value of the plant was considered for selecting the plant species.

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Materials and methods:-

Specimen collection and conduct of field trials:-

For the present investigation plant samples were collected from T.C Joseph Memorial Botanical Garden, (+10° 7' 30.65", +76° 20' 3.32"). Plant was identified by literature scrutiny, as the plant is not recorded in any local and national herbaria in South India. All field trials were conducted in the plant nursery of the Department of Botany, Union Christian College, Aluva, (+10° 7' 30.65", +76° 20' 3.32") Kerala State, India. For the sake of large scale planting material production in mechanized gardens, trials were conducted in 100cc root-trainer blocks (24 celled) using potting mixture (RTPM), root trainers with vermin-compost (RTVC) and with coir pith RTCP as rooting medium.

Experiment design:-

For all field trials, average of the three replicates during the months of June (P1), and October (P2), February (P3) was conducted in three different rooting media. Prop root cuttings with an average size of 7.5 to 10 cm were used. The sample size was kept twenty-four for each trials separately as the 100cc root-trainer block contains 24 cells. A non-auxin control and three Indole 3-butyric acid (IBA) concentrations were designed in this experiment with 300ppm, 500ppm and 1000ppm (parts per million) in order to detect the rooting/sprouting ability by quick dip method. A randomized complete block design was employed. After 45 days, the cuttings were evaluated for rooting/sprouting percentages, mortality percentage and viability percentage. The data obtained were subjected to one factor analysis, employing analysis of variance (ANOVA) and two-way ANOVA. The data generated obtained through the present investigation are being interpreted using sprouting value index (SVI) proposed by Nayagam (2015).

Results:-

Sprouting and rooting studies of prop root cuttings were carried out in three different rooting medium during three seasons in an year, for three consecutive year till 2016 and the various attributes obtained are given in table 1 to table 6. Prop root cuttings of seven-year-old *Clusia* plant was used to obtain prop root cuttings at four months interval (Fig. 1). Various attributes of SVI calculation such as CWR, SP, SUP and VP prop root cuttings in three concentrations used against the control are given (Fig 2, Fig. 3 and Fig. 4). Callus production as well as sprouting and rooting was very poor in control. High VP was obtained with all the three concentrations of IBA used (ranging between 83.05% to 95.84%) in the rooting/sprouting medium RTCP.

Fig. 1:- Rooted prop root cuttings of *Clusia fluminensis*



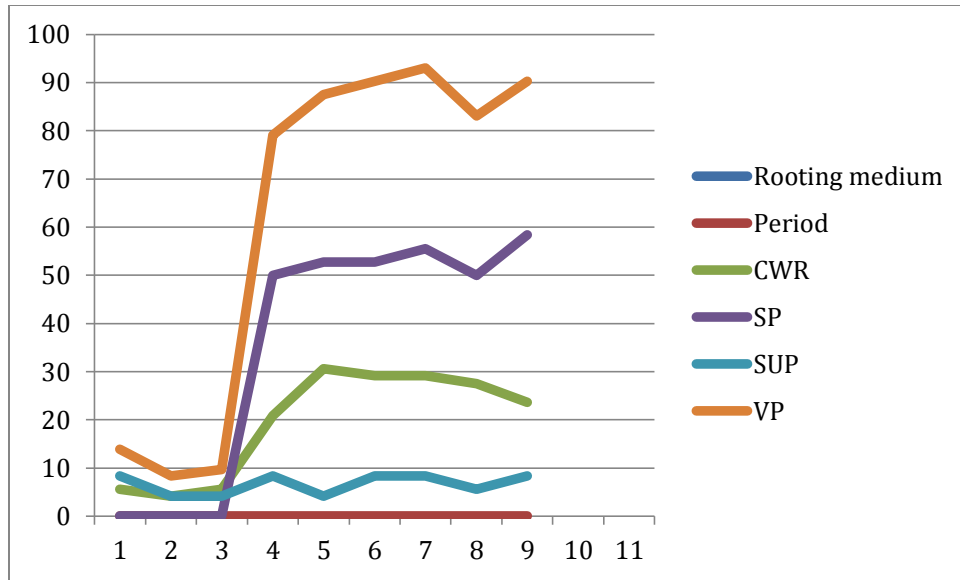


Fig 2:- Attributes of SVI in 300ppm IBA treated prop root cuttings of *Clusia fluminensis*

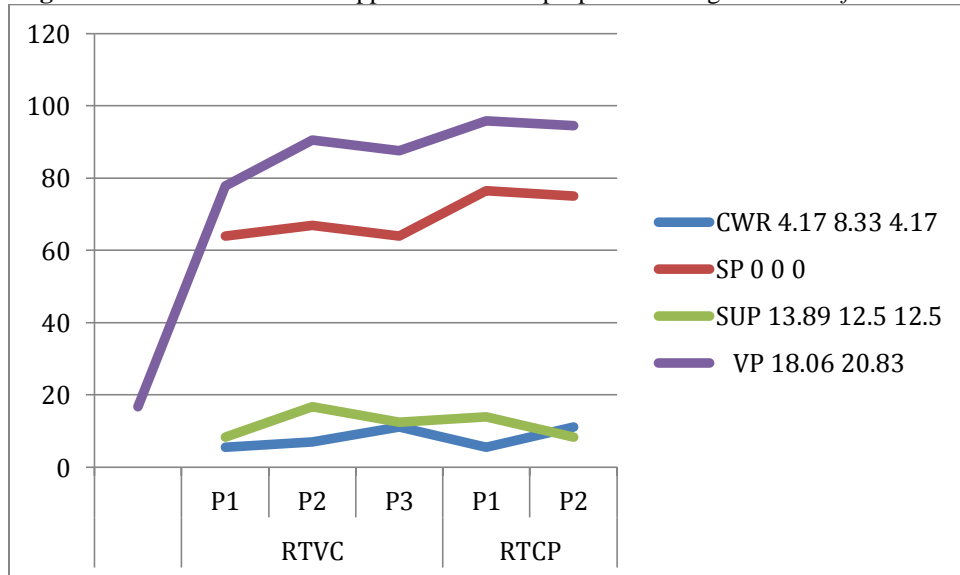


Fig 3:- Attributes of SVI in 500ppm IBA treated prop root cuttings of *Clusia fluminensis*

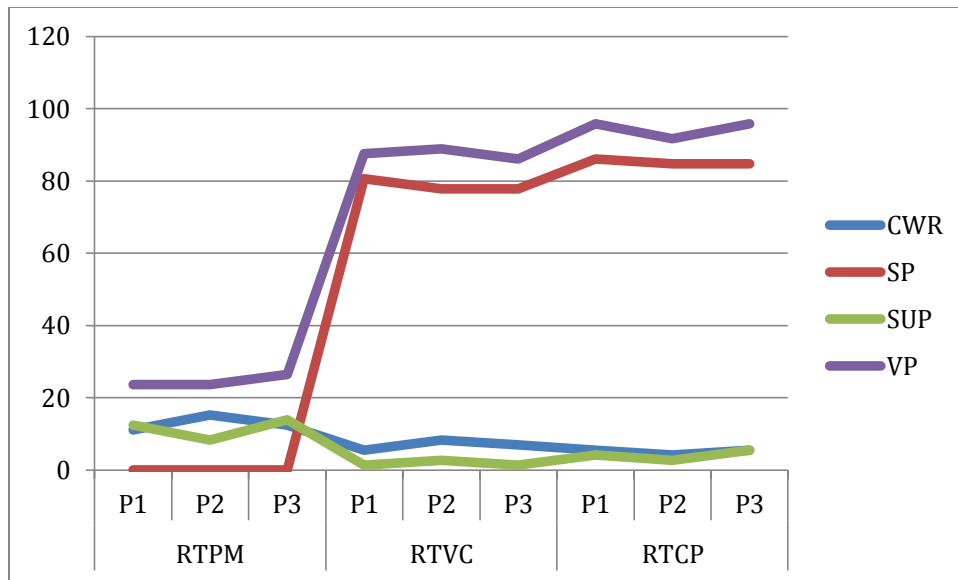


Fig 4:- Attributes of SVI in 1000ppm IBA treated prop root cuttings of *Clusia fluminensis*

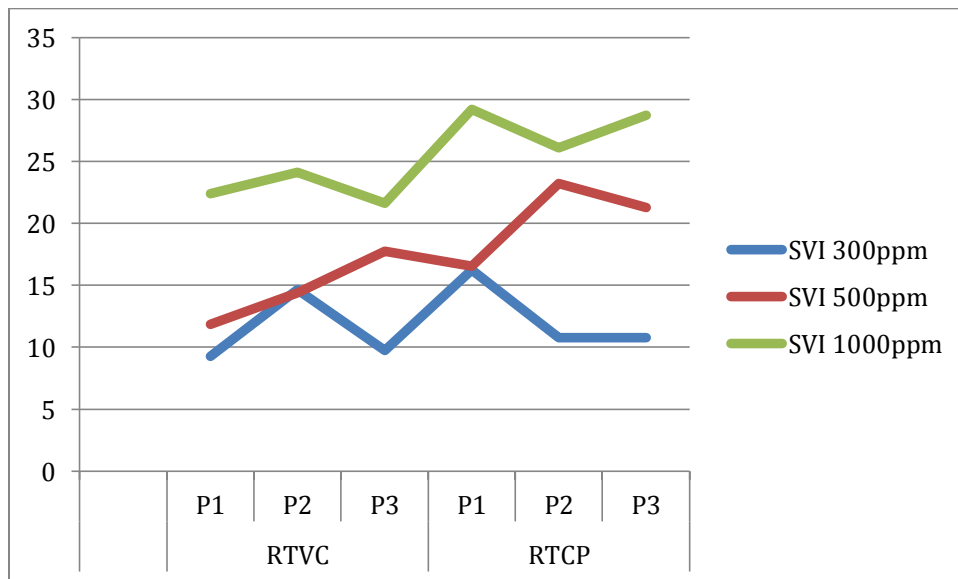


Fig. 5:- SVI results in *Clusia fluminensis* by IBA induced rooting of root cuttings

With prop root cuttings of *Clusia fluminensis*, maximum SVI was obtained in RTCP 1000ppm treated cuttings (29.17) (Fig. 5). The speed of completion of sprouting/rooting in RTVC was found lower than in RTCP. The ANOVA results on callus production show significance at 1% level between concentrations of IBA and significance at 5% level between media of study. The ANOVA results on callus formation and callus with root formation show significance at 1% level between concentrations of IBA and between media (Table 1, Table 2). The results were non significant at 5% level between years and month of study.

Table 1:- Results of ANOVA for comparing callus production on prop root cuttings of *Clusia fluminensis*

Source of Variation	Sum of Squares	Degree of Freedom	Mean Square	F-value	p-value
Medium	73.796	.1	73.796	4.092	0.049*
Year	51.627	2	25.814	1.431	0.249 ns

Month	99.721	2	49.861	2.765	0.073 <i>ns</i>
Concentration	2940.536	2	1470.268	81.528	<0.001* *
Error	829.557	46	18.034		
Total	3995.238	53			

** Significant at 1 % level; * significant at 5% level; *ns* non significant at 5 % level

Table 2:- Results of ANOVA for comparing callus and adventitious root/sprout production on prop root cuttings of *Clusia fluminensis*

Source of Variation	Sum of Squares	Degree of Freedom	Mean Square	F-value	p-value
Medium	269.128	1	269.128	38.221	<0.001**
Year	15.751	2	7.875	1.118	0.336 <i>ns</i>
Month	14.634	2	7.317	1.039	0.362 <i>ns</i>
Concentration	2979.841	2	1489.921	211.598	<0.001**
Error	323.899	46	7.041		
Total	3603.252	53			

** Significant at 1 % level; * significant at 5% level; *ns* non significant at 5 % level

The ANOVA results on callus formation and callus with root formation show significance at 1% level between concentrations of IBA and significance 5% level between media of study for callus formation root production show significance at 1% level between medium of study.

Discussions:-

ICRISAT (2007) stresses the importance of long flowering cycle of many tree species and also seed abortion (Arathi, *et al.*, 1999) in *Pongamia pinnata* as these factors influence in the reduction of germination percentage and availability of good seed samples. Production of elite genotypes of any plant species can be performed to cultivators desire by vegetative method round the year. The SVI studies worked out in the present study is a mathematical approach to determine the quality of rooting material, suitability of the medium used for rooting and the optimum concentration of IBA to be used. Production of elite genotypes of any plant species can be generated by vegetative method of propagation round the year. In field trials, all planting materials gave elite performance in RTCP planting medium. In the trials using 1000ppm IBA (Indole 3-butyric acid) treated cuttings also SVI was highest in RTCP (29.17).

Results of the experiments showed that the season, concentration of auxin (IBA) and type of cuttings were paramount importance in sprouting and rooting studies. Gurumurthi *et al.*, (1992) proved that cuttings roots more when fresh shoots start growing whereas Wareing *et al.*, (1967) were of the view that the increased production of endogenous auxin during spring might be responsible for easy rooting. Lack of rooting response during August-September and November-December may be attributed to the reduction in the endogenous level of auxin or nutritional status of the cuttings (Chauhan and Sehal, 1982; Puri and Shamet, 1988; Nautiyal, *et al.*, 1991). However in the present study all the above-mentioned problems were nullified as root cuttings do not encounter problems of reproductive cycle.

Conclusions:-

Vegetative propagation using cuttings provide genetically identical clonal plants in vast number throughout the year. The present study focuses on regeneration of plantlets from prop roots of *Clusia fluminensis* and the data obtained were interpreted using sprouting value index (SVI) to establish suitable vegetative planting material and suitable rooting medium through field trials. Results from repeated field experiments ~~Trials conducted~~ in different rooting media reveal the sprouting efficiency and vigor in different medium. Even though the age of tree together with flowering cycle and change in amount of nutrients in the vegetative parts greatly affects the rooting of cuttings; usage of stem cuttings from juvenile plants proves to be promising. This method of planting stock preparation is valuable in large-scale cultivation and is much promising in producing quality clonal planting material production from adventitious roots in economically important plants in future.

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