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Multiple Shoots from Leaf Explants of *Trichosanthes anguina L.*

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Abstract: Regeneration shoots from leaf explants of callus and In Vitro multiple shoots were obtained on MS Medium within BAP, NAA, L-Glumatic acid and Kinetin , Coconut milk (CM). High frequency plant regeneration from leaf explants of S. Nigrum by Ugander et al (2010) Callus induction and base explants of Aloe vera R. Prasad, Venkateshwarlu M et al (2018). MS basal medium supplemented with various Auxons/Cytokinins BAP and NAA.Coconut water also had a rolein triggering the formation of mult1ple shoots. Addition of BAP at 3.0 mg/l and NAA at 2.0 mg/l to the MS basal medium, induced regeneration from the leaf segments. With an increase in the level of BAP 1.0 - 3.0 mg/l the percentage of explants producing shoots also increased. The number of shoots developed on the leaf segments ranged from 1-4 to 2-3 by the addition o BAP at concentration of 1.0 mg/I or NAA at 2.0 mg/l. Among he three concentrations of coconut milk used i.e, 5, 10 and 15% of coconut milk along with 0.5 mg/l BAP proved to be ideal for multiple shoot induction. MS medium fortified with 2.0 mg/l BAP 0.5 mg/l L-2.0mgl/l Kn or 3.0 mg/l L-G Glutamic acid also induced shoot buds on leaf segments.

Keywords: Multiple shoots , Leaf Explants, NAA, L-Glutamic acid, BAP, Trichosanthes anguina L

I. INTRODUCTION

In the present paper, a simple and reproducible procedure was devised to obtain multiple shoots from leaf segments of *Trichosanthes anguina* L on MS medium fortified with plant growth regulators along with coconut milk and amino acids. The main objective of clonal propagation is to establish plants that are uniform and predictable for selected qualities. Growth or *in vitro* propagated plants is often stronger than in those cloned *in vitro* phyto chemical analysis and biological activities inmomrdica Venkateshwarlu *et al* (2011). The plants of Cucurbitaceae suffer from several diseases including the water melon mosaic virus Wayne *et al* (2011) Cucumber green mottel mosaic virus Wayne *et al* (2011) and *Solanum nigrum* also suffers from downy and powdery mildews which seriously limits the crop production. Axillary buds from pump- kin were reported by Ugender *et al* (2019) & Rathore (2010).

II. MATERIAlS AND METHODS

They were cultured on MS medium containing 2.5% sucrose and 0.8% Agar- Agar and different concentrations or BAP. NAA and L-Glutamic acid Leaf sgments of Trichosanthes were cultured and surface sterilized with 0.1% HgCl for 5-7 minutes and rinsed with sterile distilled water. Cultures were incu- bated under 16 hrs .illumination (250 lux) at 25± 2°C tempera- ture.Each treatment consisted of 10-15 replicates. The data was recorded at the end of eighth week in vitro propagation of Zyzlus Sudhershan et al (2000) cloning protocol Campstrini (2006). The pH of he medium was adjusted to 5.8 and later was autoclaved at 120 °C for 17minutes. Rajendraprasad, Venkateshwarlu M (2018) experimental mutagenesis on cicer tissue culture studies stem node explants, multiple shoots in cucumis Venkateshwarlu m (2008) and (2019).

III. RESULTS AND DISCUSSION

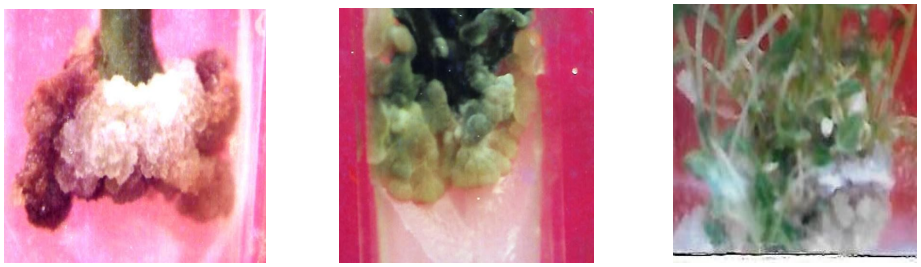
The results of the study have shown the initiation of shoot buds and formation of multiple shoots from leaf segments. Addition of NAA failedlo produces many shoots, but enlarged theleaf segments. Lower levels of coconut milk (5 & 10%) induced callus formationLeaf explants were inoculated on MS basal medium fortified with various Auxins cytokinins i.e., BAP and NAA. Coconut water also had a role in triggering the format on of multiple shoots Kanna *et al* (2005) *In Vitro* micropropagation *Solanum nigrum* Ram *et al* (2002). The mean number of shoots developed on the leaf segments ranged from 1- 4 to 2 - 3 by the additionof different concentrations of BAPand NAA the level of SAP (3.0 mg/l to 4.0 mg/l) resulted in anincrease in the percentage of shoots developed with 10, 15,20% of coconut milk also triggered the induction of multiple shoots (Plate I). Low concentration of L- Glulamic acid (0.5 - 3.0 mg/l, along with SAP (1.0 mg/l, produced significant mean number of multiple shoots that ranged from 2-3 to 5-Sin the leaf segments. Shoot multiplicalion was obtained form leaf explants cultured on MS Medium supplemented with 1.0lo 3.0 mg/I BAP.Raising the levelof BAP (0.5 to 2.0 mgn) resulted in an increase in the numberof shoots from leaf segments of *Trichosanthes anguina* L suggestedthat the forma- tion of multiple shoots at the leaf regionof the leaf of soyabean indicated the existence of totipolencyin this regionwhich can be activated with the addition of BAP. (Table-1) (Plate-I)

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Table-I Muliple shoots from leaf explants of *Trichosanthes anguina* L

Growth Regulators	Leaf Explants	
	% Frequency of Shoots	Mean no. of Shoots
MS + 0.5 mg/l BAP + NAA	**40**	**Callus**
MS + 1.0 mg/l BAP + NAA	**35**	**Callus**
MS + 2.0 mg/l BAP + NAA	**30**	**Shoots (1-2)**
MS + 3.0 mg/l BAP + NAA	**25**	**Shoots (2-4)**
MS + 0.5 mg/l NAA + Kn+CM+L-Glutamic acid	**30**	**Callus+ Small buds**
MS + 1.0 mg/l NAA + Kn+CM+L-Glutamic acid	**25**	**Callus + Small buds**
MS + 2.0 mg/l NAA + Kn+CM+L-Glutamic acid	**20**	**Shoots (3-5)**
MS + 3.0 mg/l NAA + L-Glutamic acid	**25**	**Shoots (3-4)**
MS + 4.0 mg/l NAA + L-Glutamic acid	**20**	**Shoots (4-6)**

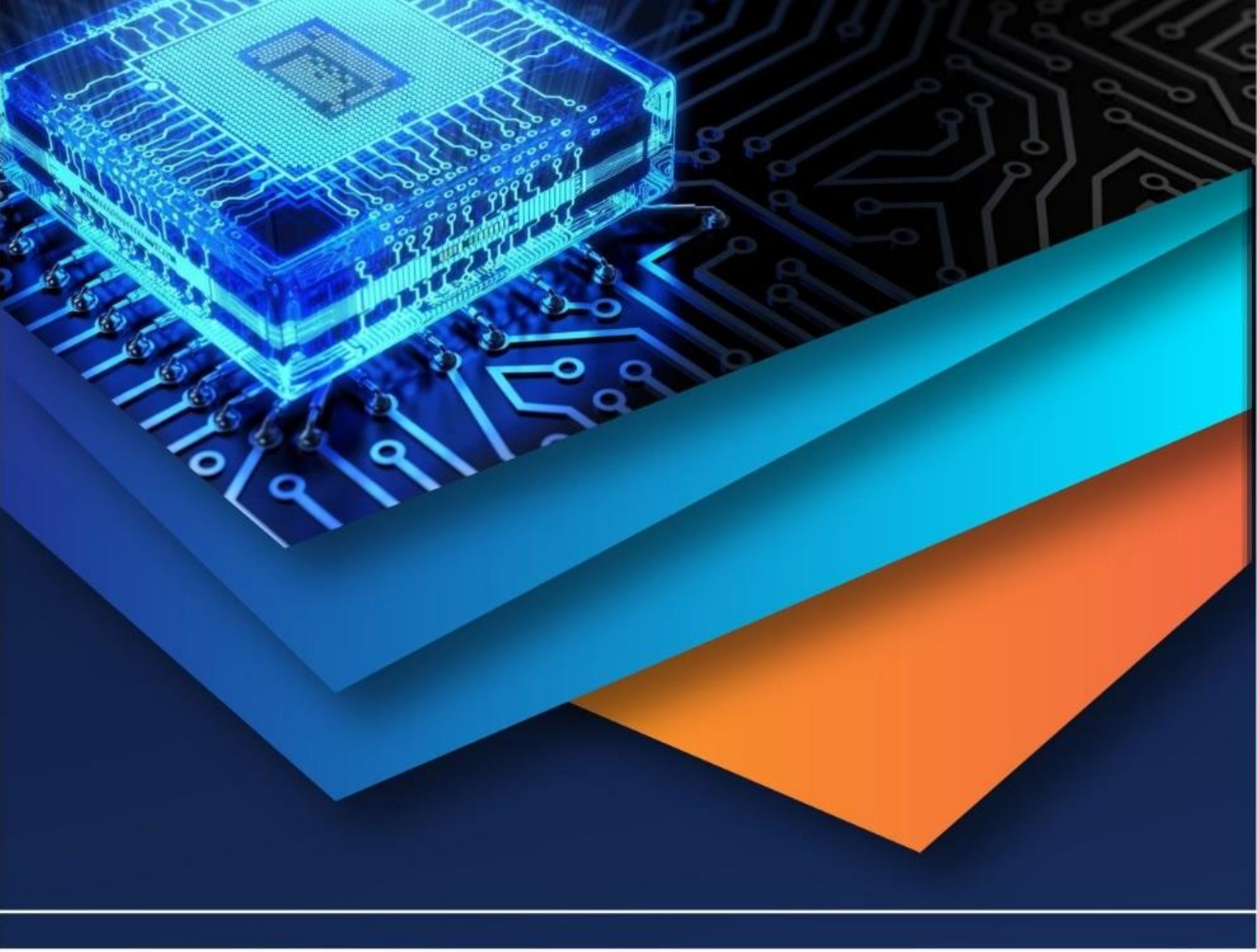
Plate-I Muliple shoots from leaf explants of *Trichosanthes anguina* L.



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