## *In-vitro* Multiplication and Induction of Microrhizome of Ginger (*Zingiber officinale* Rosc.) Cultivars Grown in Sri Lanka

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## Abstract

Ginger, Zingiber officinale Rosc. is one of the world class medicinal spices, usually propagated through the rhizome. Slow propagation rate and the risk of disease transmittance through the rhizomes have hampered the propagation. This study summarizes a method for an efficient *in- vitro* propagation and induction of microrhizome of Ginger cultivar, Local, Chinese and Rangun.

Established explants *in-vitro* were introduced to a basal Murashige and Skoog medium(MS) fortified with sixteen treatments structuring factorial combination with BAP (Benzyl aminopurine) (0, 2.0, 4.0 and 6.0 mgL<sup>-1</sup>) and NAA (Naphthalene acetic acid) (0, 0.1, 0.25 and 0.5 mgL<sup>-1</sup>) to evaluate the hormonal combinations for multiplication. Liquid and solid forms of above selected medium were evaluated for the effective multiplication. For root induction, *in-vitro* raised plantlets were cultured on full and half strength MS medium supplemented with either NAA or IBA (Indole 3 butyric acid) in concentrations of 0, 0.5, 1.0, 1.5, and 2.0 mg L<sup>-1</sup>. Rooted plants were transferred into pots containing different potting mixtures. Then these plants and the vegetative plants were established in the field. Also, experiments were conducted with different combinations of BAP and NAA, different concentrations of sucrose, the photoperiod and the solid/liquid nature of the medium to study the induction of microrhizomes of *in-vitro* produced shoots.

Local ginger showed the highest rate of multiplication in MS supplemented with  $2mg l^{-1}BAP$  and  $0.25mgL^{-1}$  NAA whereas Rangun and Chinese required 4.0 mgL<sup>-1</sup> BAP and 0.25 mg L<sup>-1</sup> NAA. The observations revealed that the liquid culture supported better multiplication though shoots in liquid media should be transferred into the solid medium to achieve better shoot growth. Local ginger showed the highest root formation in the  $\frac{1}{2}$  strength MS medium with 1.5 mgL<sup>-1</sup> while Chinese and Rangun well performed in the same media with 2.0mg L<sup>-1</sup> NAA.

The highest survival rate of 96.6% for local and 86.3% for Chinese and Rangun resulted in the potting media containing coir dust during the hardening. However the highest shoot growth was observed in the plants transplanted into the mixture of coir dust: sand: top soil: cattle manure 1:1:1:1 with a low rate of survival. In the first generation of acclimatized tissue culture derived plants in the field have shown significantly low rhizome yield compare to sucker derived plants. But *in-vitro* produced plants have shown higher rate of multiplication in three cultivars.

	Local		Chinese		Rangun	
	Tissue cultured	vegetatively propagated	Tissue cultured	vegetatively propagated	Tissue cultured	vegetatively propagated
Number of Leaves	15.68 ±0.39 <sup>d</sup>	17.4 ±0.45 <sup>bc</sup>	15.00 ±0.41 <sup>d</sup>	16.12±0.506 <sup>cd</sup>	20.3±0.86 a	18.4 ±0.37 <sup>b</sup>
Shoot height cm	47.52±0.41 <sup>d</sup>	57.4±1.004 °	75.96±1.93 <sup>b</sup>	85.36±0.82 ª	77.56±1.87 <sup>b</sup>	60.16±2.65 °
Number of shoots	90.8±4.75 <sup>a</sup>	19.52±0.58 <sup>d</sup>	44.87±1.52 °	14.56±0.20 <sup>d</sup>	67.8±1.82 <sup>b</sup>	19.4±0.65 <sup>d</sup>
Fresh weight of shoots g	1114.6±100.3ª	263.932±6.8 <sup>b</sup>	1243.4±56.67ª	277.21±10.2 <sup>b</sup>	1097.6±61.18 ª	301.58±11.22 <sup>b</sup>
Fresh weight of rhizome g	649.00±22.65 °	676±29.9°	838±81.5 <sup>b</sup>	1220.48±82.2ª	858.2±30.4 <sup>b</sup>	1307.46±27.5ª
Girth cm	0.56±0.008 <sup>a</sup>	0.807±0.02 <sup>a</sup>	0.60±0.01 <sup>b</sup>	0.806±0.008ª	0.57±0.02 <sup>b</sup>	0.77 0±03 <sup>a</sup>
Fresh weight of roots g	37.4 ±3.95 <sup>ab</sup>	38.4 ±1.91 <sup>a</sup>	8.6±0.98°	32.6±1.32 <sup>ab</sup>	10.6 ±2.22 <sup>c</sup>	31.00 ±3.5 <sup>b</sup>

Table: Growth performances of tissue cultured and conventional propagated ginger in the field

Values are mean followed by the same letter(s) are not significantly different at p=0.05

The weight of microrhizome significantly increased in Local and Rangun up to 4 mg  $L^{-1}$  of BA with 0.1  $L^{-1}$ NAA whereas Chinese well performed in 0.1mg $L^{-1}$  NAA with 6mg  $L^{-1}$  BAP. This experiment proved the higher concentration of BAP inhibits the increase of weight in microrhizomes of Local and Rangun. MS medium with 90g $L^{-1}$  sucrose showed the optimum performances in induction of microrhizomes of the three cultivars with the highest and the lowest weight were observed respectively in Rangun and local. Concentration of sucrose from 90g  $L^{-1}$  to120 g $L^{-1}$  induced a high number of small shoots while fresh and dry weight of the rhizome were decreased. Different photoperiod exposure levels revealed that 16 hrs of light and 8 hrs of dark condition with solid medium produced the highest fresh weight of microrhizomes. Further studies are necessary to evaluate the rhizome yield and growth characteristics of second generation. The procedure developed in this study will be useful for rapid *in-vitro* propagation and production of healthy microrhizomes of ginger.

Keywords : Ginger; Explants of sprouted buds; Micropropagation; Zingiber officinale

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