Cost effective mass multiplication of *Gymnema sylvestre* in hydroponic system

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Abstract

Present experiment was designed with an objective to develop a cost efficient mass multiplication method for *Gymnema sylvestre* using hydroponic system. A plastic tub, with polyethylene cover, containing 1/10 strength of MS salts supplemented with Indole butyric acid (IBA) at different concentrations (0.5, 1.0, 2.5 mg/L) was studied. Medium containing 0.5mg/L of IBA produced highest rooting (66%) with 96 % survival. This protocol will serve as an alternative to the existing *in vitro* and clonal multiplication protocols.

Key words: Hydroponic, Multiplication, Rooting, IBA, *Gymnema sylvestre*

Introduction

Gymnema sylvestre R.Br. (Asclepiadaceae), a perennial plant, stout woody climber with long slender branches is distributed throughout India, in dry forests up to 600 meter height. The leaves of this plant are opposite, entire, 1 to 3 inches long, and 1 to 2 inches broad, elliptic or obovate, acute or cuspidate, rarely cordate at the base, membranous, thinly pubescent on both sides, the upper surface often darker green than the lower[1]. As it is one of the important anti-diabetic medicinal plants there is a growing demand for *G. sylvestre* leaves in the pharmaceutical trade. Gymnemic acid, the active principle of this plant, is extracted from leaves and used widely as anti-diabetic[2], anti-sweetner[3], and antihypercholesterolemic[4]. Due to indiscriminate collection and over exploitation, natural stands of *G. sylvestre* are fast disappearing and threatened. Hence to avoid its disappearance, it requires cultivation in farmlands and this approach offers excellent scope for cultivation in the subtropical regions in southern India. Conventional propagation of this plant is hampered due to poor seed viability, low rate of germination, poor rooting ability of vegetative cuttings and low multiplication rate even in tissue culture[5]. Reports of rooting of *G. sylvestre* using IBA in tissue culture technique was also very low[6, 7]. Hydroponics offers opportunities to provide optimal conditions for plant growth and enables the growers to manage the supply of essential nutrients to crops more efficiently and accurately than traditional field systems[8]. Hydroponic system has been developed for growing several plants[9-12].

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Therefore, hydroponic system was looked at as a potential alternative for rooting and mass multiplication of *G. sylvestre*. No report on hydroponic system based rooting of *G. sylvestre* has been published so far. MCRCGY1, an accession collected from Muniyankudisai Village, Tamilnadu was micropropagated using MS basal medium supplemented with Benzylaminopurine (BAP - 3.5 mg/L), Kinetin (KN 1.0 mg/L), Napthaleneacetic acid (NAA 0.2 mg/L) and the grown plantlets were established at Sri AMM Arunachalam Technology Resource centre, Vadakadambadi, Tamilnadu.

Material and methods

Young stem cuttings with an actively growing side branch obtained from 20 months old tissue culture raised plants were used as explants. Explants with different lengths viz., 10.0 cm, 30 cm and 45 cm were placed in Thermocole sheets (10 mm) with an internal spacing of 5x5 cm. The cut ends of the planting materials along with the Thermocole sheets were dipped to a depth of 1.0 cm in 20 liters of nutrient solution containing 1/10 Murashige and Skoog (MS) salts[14] in water with different concentrations of IBA (0.5, 1.0, 2.5, mg/l) in plastic tubs (4 x 2 feet). The tubs were tightly covered with polythene sheet. The final pH and electrical conductivity of the solution were 5.9, and 859.9 µS. The nutrient solution was changed once in seven days. The diurnal temperature fluctuated between 28^0C to 34^0C during the study period. Each treatment had 12 replicates and the experiment was repeated twice. Data were recorded after 21 days of culture.

Results and Discussion

Preliminary studies showed that the explants with an actively growing side branches responding better than those actively growing single shoots and leaf less stem cuttings. Results of the present study indicate that both planting material size and IBA concentration play

a major role in the induction of roots in *G. sylvestre*. 1/10 MS basal salts medium supplemented with IBA induced better rooting when compared to control (Table 1, Fig 1). Root initiation was observed within a week of incubation and from second week onwards they were more visible. Among the different size of planting material used, the 30 cm long explants exhibited better root induction (93% rooting, 17.8 ± 1.1 root numbers, and 15.4 ± 0.8 cm root length) on 1/10 MS salts supplemented with 0.5 mg/L IBA followed 1.0 and 2.5 mg/L IBA and control. 1/10 MS medium and IBA have been reported to prevent leaf fall and promote shoot growth[15].

Table 1. Rooting of *G. sylvestre* stem cuttings grown under hydroponic system with 1/10 MS salts supplemented with different IBA concentration

Treat.	Explant size (cm)	Results*		
		Rooting (%)	No. of roots / explants **	Root length (cm) **
Control	10	50	7.3 ± 0.5^{cd}	9.2 ± 0.5^{cde}
	30	66	11.5 ± 0.5^{b}	12.6 ± 0.3^{b}
	45	33	3.5±0.7 e	7.2 ± 0.5^{def}
IBA (mg/L)				
0.5	10	66	9.7 ± 0.9^{bc}	11.9 ± 0.5^{b}
	30	93	17.8 ± 1.1^{a}	15.4 ± 0.8^{a}
	45	50	10.3 ± 1.5^{b}	9.7 ± 0.4^{c}
1.0	10	66	9.5 ± 0.5^{cd}	7.5 ± 1.0^{cde}
	30	83	15.6 ± 1.1^{a}	8.1 ± 1.1^{cd}
	45	50	9.3 ± 0.5^{bc}	5.8 ± 1.3^{ef}
2.5	10	50	6.3 ± 0.5^{e}	9.0 ± 0.4^{cd}
	30	83	11.4 ± 1.6^{b}	8.6 ± 1.7^{cd}
	45	33	5.5 ± 2.1^{de}	5.2 ± 0.8^{f}

* - Results were recorded after 21 days. Control: 1/10 MS salts without hormones.
** - Values are mean ± SD, $n = 12$; means for each experiment marked with same letter do not differ significantly ($p<0.05$).

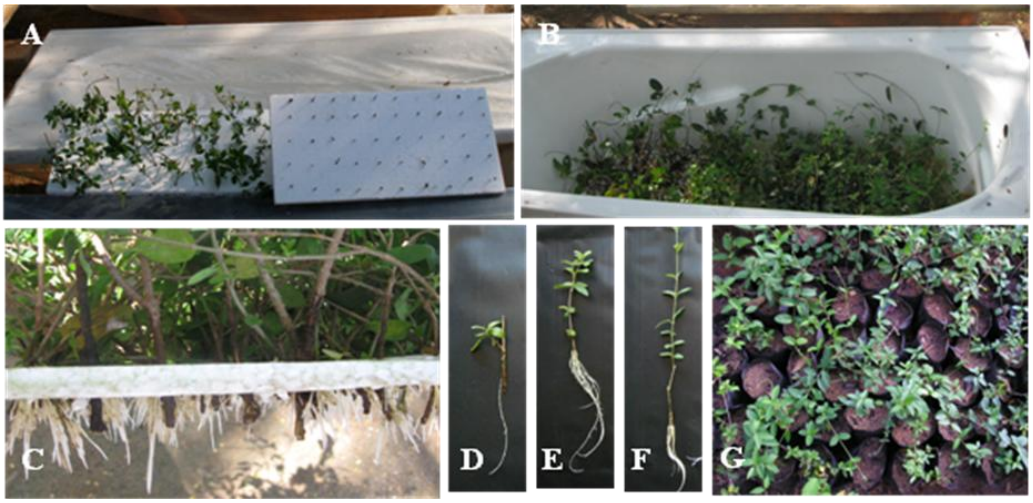

Fig. 1: A - Hydroponics setup for growing *G. sylvestre*, B, C 14 days old *G. sylvestre* explants and rooting, D, E, F - 10 cm, 30 cm and 45 cm long explants after 21 days, G – Hardened plants in poly bags

Hardening of plantlets was achieved in poly house with red soil, river sand, farm yard manure (1:1:1) with 96% transplantation success. With this protocol more number of plantlets can be produced within 21 days. As this is the first report on soilless cultivation of *G. sylvestre* stem cuttings, using hydroponic system for mass propagation, use of plastic tanks with nutrient solution forms a very attractive concept. Further studies on continuous cultivation and evaluation of secondary metabolite production will throw more light on the superiority of this soil less cultivation technique than conventional systems.

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