In vitro PROPAGATION OF *Gymnema sylvestre* R.Br. (MASBEDDA)

BY

MOHIDEEN HILAL MOHAMMED ALI

Department of Agricultural and Plantation Engineering
Faculty of Engineering Technology
The Open University of Sri Lanka
Nawala, Nugegoda
Sri Lanka
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ABSTRACT

Gymnema sylvestre is an endangered medicinal plant and red listed in the IUCN guidelines. The plant has increased its uses since recently due to the pharmaceutical potential of Gymnemic acid, found in the leaves. *G. sylvestre* is conventionally propagated through seeds but the attempt for its production and conventional propagation is hampered due to its seasonal bearing habit, rareness of the plant and the loss of seed viability quickly. Thus efficient *in vitro* plant production system for *G. sylvestre* is required to produce enough true-to-type, uniform quality, disease-free planting materials. Therefore, this research project was undertaken to develop a micropropagation protocol for *G. sylvestre* using seedlings. Stored seeds (3 months in refrigerator) were cultured on Murashige and Skoog (MS) medium treating with commercial clorox (5% NaOCl), 10%, 20%, 30% concentrations with 10, 15, 20 minute exposure times to identify the best surface sterilization procedure. After 30 days, germination percentages and non contamination percentages were recorded to identify the best clorox concentration and exposure time for surface sterilization of seeds. Seeds were sterilized using the best clorox concentration and exposure time and cultured on Woody Plant Medium (WPM) and MS basal medium to find out the best medium for seed development. After 42 days of observation axillary buds were excised from the basal medium and transferred to WPM medium with different concentrations of 6-benzylamino-purine (BAP) (2.5, 3.0 mg/L), α- napthaline acetic acid (NAA) (0.1, 0.2 mg/L) as a combination to identify the best phytohormone combination for *in vitro* proliferation. Axillary buds were excised at 42 days and transferred to WPM media containing different concentrations of Indol Butric Acid (IBA) (0, 1, 2, 3 and 4 mg/L) with and without activated charcoal (1g/L) to form roots. All the experiments were conducted according to Completely Randomized Design (CRD) with 20 to 60 replicates. Data were collected in two week intervals and analyzed using SAS & Minitab computer softwares.

Clorox (5% Naocl), 20% concentration with 15 minute exposure time found to be the best surface sterilization procedure recording the highest germination percentage (40%) and highest non contamination percentage (100%) on MS basal medium within 30 days. WPM basal medium was superior to MS basal medium, which gave a higher germination percentage (63%) and higher shoot height (3.51 cm) at 42 days. WPM medium with 2.5 mg/L BAP, 0.1mg/L NAA produced 2.92 mean number of shoots per axiliary bud with 2.13 cm mean shoot height and 7.08 mean number of leaves after 42 days. IBA 4 mg/L without activated charcoal showed a higher root length and higher number of roots compared to the treatment combinations with 1 g/L activated charcoal was used.