CULTIVATION OF *Munronia pinnata* (Binkohomba): SOME ASPECTS OF MICROPROPAGATION, AGRONOMIC PRACTICES AND PHYTOCHEMICAL SCREENING

By

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ABSTRACT

Munronia pinnata (Wall) Theob, (Sinhala- Binkohomba) is a threatened medicinal plant of the family Meliaceae. It is a small, hardy, perennial plant naturally growing in the dry, dry- mixed to wet evergreen forests and some savannah lands in Sri Lanka. The entire plant is used for the medicinal purposes. Due to the large scale and unrestricted exploitation of natural habitats to meet its ever-increasing demand coupled with insufficient natural regeneration, the wild stock has been markedly depleted. Large-scale cultivation having the quality of wild grown plants is an alternative for conservation and sustainable use of this threatened species.

Studies were conducted to determine the effect of light, planting spacing and fertilizer types on growth, biomass production and chemical composition of *M. pinnata* during the two main cultivation seasons (*Yala* and *Maha*) in low country dry zone of Sri Lanka. Since fertilizer application is necessary in long term cultivation programs, a parallel soil fertility analysis was done in both seasons.

In *Maha* (2004), 100 % light level indicated an adverse effect on growth and biomass production of *M. pinnata* in field cultivation. In *Yala* (2005) plants were cultivated only under 10 % sunlight. In both seasons, application of inorganic fertilizer resulted in a significantly higher ($p=0.05$) above ground and below ground biomass production of *M. pinnata* at the harvesting stage. The growth rate of *M. pinnata* was not affected by the type of fertilizer though inorganic fertilizer treatment induced better growth. On per plant basis, better growth of *M pinnata* were recorded at harvesting stage of inorganic fertilizer treatment with 50 x 50 cm spacing in both *Maha* (2004) and *Yala*

(2005). The chemical composition of cultivated and wild grown plants, in the same stage were compared in both *Maha* (2004) and *Yala* (2005) seasons using Thin Layer Chromatography (TLC). TLC chromatograms of wild and cultivated plants were similar, suggesting that the fertilizer application (organic or inorganic) and plant spacing (25 x 25 cm or 50 x 50 cm) do not change the chemical composition of *M. pinnata* cultivated under 10 % light intensity.

Cattle manure added less N, P and K, than the inorganic fertilizers. The application of organic or inorganic fertilizer affected soil chemical properties across both *Maha* (2004) and *Yala* (2005) seasons under *M. pinnata* cultivation. At the end of each cropping season, negative balance of N, P and K was recorded for most treatments. Although the soil nutrient content depends on many factors, the data support the notion that application of fertilizer is important in long-term cultivation of *M. pinnata* to keep the positive nutrient balance at the beginning of the cropping season.

The study also established a tissue culture protocol for achieving high-frequency axillary shoot multiplication and direct organogenesis for large-scale production of *M. pinnata* and to compare chemical composition of wild -grown plants and *in vitro* grown plants.

In the nodal culture, MS medium supplemented with 2.00 mg/L BAP showed the highest shoot initiation and BAP concentration of 5.00 mg/L negatively modified the frequency of shoots induction and growth. The maximum percentage (70 %) of shoot generating explants were observed on MS medium with 1 mg/L BAP and 2 mg/L IBA, which induced the highest average number of shoots (8.55 ± 1.47). Rooting of

plantlets was observed in 8 weeks of culture in MS medium supplemented with different combinations of auxins and sugar percentages. IBA (1 mg/l IBA) was more suitable for root induction resulted the highest percentage of root induction (90 %) together with the highest root number (4.2 ± 0.66). Rooted shoots were subsequently transplanted to small pots filled with sand and were acclimatized. Plantlets could be successfully established in soil with a high survival rate (80%) after eight weeks in the green house. The regenerated plants did not show detectable variation in morphological or growth characteristics. The TLC chromatogram pattern in the cloned plants were identical to that of the mother plant, under both 253 nm and 366nm wavelength for all extracts, indicating that the process of regeneration had not altered chemical composition of *M. pinnata.*

Overall, the present study provide the evidence and support for the possibility of large-scale field cultivation of *M. pinnata,* named under 50 most important medicinal plants in Sri Lanka thus answering a question of national interest.

Key words: *Munronia pinnata, Yala, Maha,* fertilizer application, plant spacing, Chemical composition, Thin Layer Chromatography, nodal culture, leaf culture