T2-P13

Establishment of Cell and Root Suspension Cultures from *Curcuma xanthorhiza* for the Production of Xanthorhizol

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This research aims to establish a cell suspension protocol for *Curcuma xanthorhiza* (temulawak) as an alternative source of the bioactive compound xanthorhizol. The prime advantages of this method is that the callus can be maintained in a controlled environment for sustainable production of xanthorhizol and also to be less dependant on the mother plant. In addition, superior lines could be selected to ensure high yielding cell lines. *C.xanthorhiza* is known to produce of a number of secondary metabolites with potential applications in healthcare, pharmaceutical and cosmecuetical industries.

Tissue culture technique has enabled the development of an alternative approach for the production of these plant biochemicals of which one such compound is xanthorhizol. For the callus culture protocol, shoot buds from sprouted rhizomes were used as explants. These shoot buds were then sprouted in soil-free conditions to avoid contaminants from the soil. The usual practice of using antibiotics to suppress soil microbial contaminants was found to be not particulary suitable for *Curcuma xanthorhiza*. Callus were subsequently initiated from small and active buds of the rhizomes, sliced and cultured on MS basal medium supplemented with various combinations of phytohormones under dark condition. After 4-8 weeks, they were transferred to M2D media for cell suspension maintenance. Subculturing of the cell suspension into fresh M2D was performed every fourteen (14) days. For root cultures, the same callus was transferred to MSO liquid media. The solvent extracts from the roots and cells were analyzed using capillary GC and GC-MS. The results showed the presence of xanthorrhizol in these extracts. Interestingly, the amount of xanthorhizol was comparatively higher in both cells and root cultures compared to the culture media.