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Title: **HPLC-Based Elucidation of Tannins from the Tissue and Callus Culture Extracts of Selected Medicinal Plants**

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HPLC-Based Elucidation of Tannins from the Tissue and Callus Culture Extracts of Selected Medicinal Plants

Abstract

Background: Optimized HPLC profiling is a powerful and effective analytical tool to standardize plant samples and authenticate plant materials. In this study, three selected medicinal plants namely: *Achyranthes aspera*, *Ipomoea hederacea,* and *Ocimum basilicum* were subjected to callus induction following seedling, leaf, and stem germination.

Materials and Methods: The induced callus was subsequently dried, finely ground, and extracted using methanol and water for HPLC analysis. A validated procedure was employed to identify and separate the tannin content in seedling leaf, stem, and callus culture extracts. HPLC fingerprinting was performed using a Shimadzu LC-20A system with a retention time of 2.9 minutes at 270 nm. The aim was to ensure quality and consistency in tannin analysis across different plant parts and callus culture samples.

Results: The highest callogenic response occurred in A. aspera leaf explants on MS medium with 2.0 mg/L 2,4-D and 4.0 mg/L NAA, producing green, granular callus. The lowest was in *I. hederacea* stem explants with 0.5 mg/L 2,4-D and BAP, yielding brown, granular callus. *O. basilicum* leaf callus extract showed the largest sample area (9365.56) and tannin content (2.66), with superior precision in tannin analysis for O. basilicum and A. aspera (7.81).

Conclusion: HPLC profiling proved to be an accurate, efficient, and precise method for evaluating tannin content in selected plant samples. It is a crucial method to standardize the quality of medicinal plant compounds.

Keywords: analytical tool, callus culture, explants, HPLC, medicinal plant