**The effect of plant growth regulators on the antioxidant enzyme activity and secondary metabolite production in the cell suspension cultures of *Melia azedarach* L.**

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**Abstract**

*Melia azedarach* L. from the Meliaceae contains a wide range of secondary metabolites used as antibacterial, antiviral, antioxidant, and anticancer in traditional medicine. The results showed that the inflorescence and petiole explants had a high percentage of callus induction compared to the leaf explants, whereas the highest callus growth was obtained from the leaf explants cultured on the MS medium containing 5 mg l-1 Naphthaleneacetic acid (NAA) and 5 mg l-1 [Benzylaminopurine](https://www.plantcelltechnology.com/benzylaminopurine-bap-solution/) (BAP) and 5 mg l-1 NAA and 3 mg l-1 kinetin Kin. Furthermore, the highest cell growth was obtained from the leaf and inflorescence callus transferred to liquid MS medium supplemented with 1 mg l-1 [2,4-dichlorophenoxyacetic acid](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5101310/) (2,4-D)+1 mg l-1 Kin and 1 mg l-1 BAP. The highest activity of antioxidant enzymes and the lowest protein content was related to the leaf cell suspension cultures in liquid MS medium containing 3 mg l-1 NAA and 3 mg l-1 BAP. However, the highest amount of proline was observed in the inflorescence cell suspension cultures in liquid MS medium containing 3 mg l-1 NAA and 3 mg l-1 Kin. The cell suspension cultures derived from inflorescence callus in MS medium containing 3 mg l-1 NAA+1 mg l-1 BAP exhibited the highest rutin accumulation (47.536 mg g-1 FW). However, the highest amount of quercetin (8.570 mg g-1 FW) and kaempferol (5.420 mg g-1 FW) accumulation were obtained from the cell suspension cultures derived from the petiole explant in the MS medium with 1 mg l-1 2,4-D+1 mg l-1 Kin. These results will facilitate the exploitation of in vitro plant cell cultures to produce high-value secondary metabolites.

**Keywords:** Antioxidant enzymes, In vitro culture, inflorescence, Kaempferol, *Melia azedarach* L., Quercetin, Rutin, Secondary metabolites

**Figure S1-** The standard curve of protein by different concentrations of bovine serum albumin

**Figure S2-** The standard curve of different concentrations of H2O2

**Figure S3-** The standard curve of different concentrations of L-proline

**Figure S4-** The standard curve of different concentrations of gallic acid

**Figure S5-** The standard curve of different concentrations of quercetin in methanol

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| **a** |
| **b** |
| **c** |
| **Figure S6-** The standard curve of different concentrations of rutin (a), quercetin (b), and kaempferol (c) by HPLC |