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Green Synthesis of Methyl Gallate Conjugated Silver Nanoparticles: A Colorimetric Probe for Gentamicin

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Extraction and isolation of methyl gallate (MG) from C. pulcherrima pods

The fresh, undried and uncrushed pods (7.7 kg) of *C. pulcherrima* were extracted with methanol for three times at room temperature. The extracts were filtered and evaporated on rotary evaporator giving the residue, CP-Pod-M (610 gm) which was dissolved in water, and subjected to liquid-liquid extraction with petroleum ether (PE) followed by ethyl acetate (EA). Ethyl acetate (EA) phases were washed with water dried with sodium sulphate anhydrous, filtered and evaporated at reduced pressure giving the residue CP-Pod-EA (70.7 g). A small quantity (3.5 g) of CP-Pod-EA was fractionated by solvent-solvent separation by using different ratios of PE:EA (40:60 \rightarrow 50:50, 10 fractions) mixture, which led to the isolation of a pure compound in one of the above fractions (PE:EA 1:1) (Rf= 0.6, EA: MeOH 9:1, silica gel 60 F₂₅₄). It was identified as methyl gallate (MG) through spectral studies (¹H NMR, EI-MS, UV and IR spectroscopy). The structure was confirmed through comparison of its TLC profile and spectral data with the authentic sample of MG.

Characterization of methyl gallate (MG)

State: White powder, $R_f = 0.6$ (EA : MeOH, 9 : 1, Silica gel), EI-MS m/z (%): 184 (M⁺, 89), 153 (100), 125 (28), 107 (7), 79 (10). ¹H-NMR (δ) (300 MHz, C₃D₆O): 3.78 (3H, s, OCH₃), 7.11 (2H,s, H-2, 6), 8.14 (3H, s, 3OH, disappeared upon D₂O shake). IR (CHCl₃) υ_{max} 3409, 3303, 2954, 1676, 1612, 1440, 1311, 1259 cm⁻¹; UV (MeOH) λ_{max} : 272, 268, 261, 225, 206 nm.



Figure S1. UV-visible spectra of MG-AgNPs indicating Stability of MG-AgNPs as a function of residence time at ambient conditions



Figure S2. UV-visible spectra of MG-AgNPs indicating electrolyte effect on **MG-AgNPs** with various salt concentrations (1mM-2M).



Figure S3. UV-visible spectra of MG-AgNPs after boiling at 100 °C for 30 minutes



Figure S4. UV-visible spectra of MG-AgNPs indicating effect of pH on stability of MG-AgNPs



Figure S5. Structures of methyl gallate, gentamicin and other competitive drugs used in this study



Figure S6. Atomic force micrographic images of, A) MG-AgNPs; B) MG-AgNPs/gentamicin complex.



Figure S7. Size distribution of the GM-AgNPs, A) Before addition of Gentamicin, B) After addition of Gentamicin



Figure S8. Job plot for binding ratio