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Tips and Tricks: Antibacterial Assay of Plant Extracts

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ABSTRACT

Antibacterial study of plant origin has brought tremendous breakthrough in various field vis. medicinal, pharmaceutical and food preservatives. The long and tedious process of identifying the capability of extracts due to the need of selecting of smaller sample particle size, maturity of sample, matrix interference, appropriate solvent polarity, substrate to solvent ratio, extraction technique, and sample storage and temperature promises high turnout of antibacterial capacity. Other consideration for instance extraction temperature, duration time, added process such as sonication, sample pretreatment affect the antibacterial extraction. Solvents used in extraction have bactericidal effect on pathogens tested. Agar diffusion and broth dilution are endpoint methods while descriptive methods involve turbidity assays and inhibition curves are used in antibacterial evaluation. Broth dilution method by spectroscopy instrument involves microscale and macroscale volume up to 250 μ L and 1 mL respectively. Soluble concentration equivalent to lower concentration of non-polar extract reduces effect of precipitation in minimum inhibitory concentration (MIC) test. MIC is easily determined by polar extracts since it is immiscible with dimethyl sulfoxide (DMSO) and broth media. Minimum bactericidal concentration (MBC) is identified at higher concentration than MIC. Determination of MIC₀, MIC₅₀ and MIC₁₀₀ can be obtained from turbidity assays. Lethality of pathogens can be established at concentration lower than MIC through inhibition on profile curves. Each test should include negative (solvent and DMSO) and positive (penicillin or tetracycline) measures to support the data analysis. Complementary antibacterial assays are recommended for confirmation of antibacterial properties from plant extracts.

Keywords: *antibacterial assay, plant extracts, minimum inhibition concentration, minimum bactericidal concentration, inhibition profile curve.*