

The Efficacy of Endophytic Fungus of *Mentha Piperita* Origin as An Antibacterial Agent

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Structured Abstract

Background: The rise of multidrug-resistant bacteria, including Methicillin resistant *Staphylococcus aureus* (MRSA) poses significant challenges to public health due to their ability to develop resistance to antibiotics making infections difficult to treat. This study focuses on the antibacterial properties of the endophytic fungus isolated from *Mentha piperita* (peppermint), a plant that is known for producing essential oils with therapeutic properties. This study aimed to investigate the efficacy of fungal extracts as antimicrobial agents against pathogenic bacteria.

Methods: Endophytic fungi were cultured, maintained and fermented for 14 days in potato dextrose broth. The fermented fungal broth was filtered and extracted using ethyl acetate solvent. The upper organic layer was collected and concentrated using rotary evaporator until the fungal crude paste was obtained. The antibacterial activity was tested against pathogenic bacteria including Gram-positive and Gram-negative bacteria by using disc diffusion susceptibility assay. Minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) were performed to determine the lowest concentration of fungal extract required to inhibit and kill bacterial growth. ATR-FTIR analysis was employed to compare the chemical differences between bacteria untreated and treated with the fungal extract.

Results: The fungal crude extract exhibited strong antibacterial activity against both Gram-positive and Gram-negative bacteria. *Staphylococcus aureus* is the most susceptible with the highest inhibition zone was observed at 20.3±0.5 mm. This inhibition zone was larger than those produced by Chloramphenicol. No inhibition zone was observed for *Pseudomonas aeruginosa* may be due to its cell wall structure that has lipopolysaccharide (LPS) which act as a barrier to antimicrobial agent which make it highly resistant. MIC & MBC values of ethyl acetate fungal crude extract on Gram-positive bacteria ranging from 1.563-3.125 µl/mL and 1.563-12.50 µl/mL respectively, whereas the MIC & MBC value for Gram-negative bacteria in range of 1.563-3.125 µl/mL and 3.125-6.250 µl/mL respectively. The extract exhibited both bactericidal and bacteriostatic effects with the MBC/MIC ratio ≤4 and >4 respectively.

Conclusion: The study revealed that the fungal extract possesses antimicrobial activity against Gram-positive and Gram-negative bacteria. All Gram-positive bacteria were susceptible to endophytic fungal extract and the extract was active against all Gram-negative bacteria except *Pseudomonas aeruginosa*.

Keywords: *Mentha piperita*, Endophytic fungus, Antibacterial activity, Gram-positive, Gram-negative

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