COMPARATIVE METABOLOMIC ANALYSIS OF SECONDARY METABOLITES PRODUCED BY ENTOMOPATHOGENIC FUNGI

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Entomopathogenic fungi, microorganisms that infect and seriously disable or kill insects, are widely recognized for producing bioactive secondary metabolites with significant biotechnological potential. In this study, we investigated the chemical profiles of three entomopathogenic fungi, *Beauveria bassiana*, *Metarhizium anisopliae*, and *Purpureocillium lilacinum,* by analyzing the production of intracellular and extracellular metabolites using liquid cultivation in Potato Dextrose Broth (PDB). Intracellular metabolites were extracted from fungal biomass using a methanol:water (4:1, v:v) solution, while extracellular metabolites were isolated from the culture supernatant by liquid-liquid partitioning with ethyl acetate. Metabolomic analysis was conducted via liquid chromatography-tandem mass spectrometry (LC-MS/MS), following by data processing in the software MzMine and molecular networking analysis performed in the Global Natural Products Social Molecular Networking platform (GNPS). Results revealed distinct metabolomic signatures across species and extraction types. *Metarhizium anisopliae* was characterized by destruxins (A, B, E1, B1), known for insecticidal and immunosuppressive properties. *Beauveria bassiana* displayed a profile dominated by beauvericin, alkaloid derivatives, and diketopiperazines., *Purpureocillium lilacinum* exhibited significant biological activities, according to the literature. Extracellular extracts showed greater chemical diversity than intracellular extracts, suggesting secretion as the primary route for metabolite release. These findings highlight the metabolic diversity of entomopathogenic fungi and the effectiveness of LC-MS/MS and molecular networking for profiling secondary metabolites, providing a foundation for further studies on purification, structural elucidation, and bioactivity for biotechnological applications.

Keywords: *Beauveria bassiana*, *Metarhizium anisopliae*, *Purpureocillium lilacinum, LC-MS/MS, molecular networking. natural products*