

ISOLATION OF ERGOSTEROL PEROXIDE WITH ANTIMICROBIAL POTENTIAL FROM DIAPORTHE HONGKONGENSIS

Andrei da Silva Alexandre^{1,2*}, David Ribeiro da Silva¹, Cecilia Veronica Nunez^{1,2}

and.alexandre36@gmail.com

1-Laboratório de Bioprospecção e Biotecnologia, INPA, Av. André Araújo, 69067-375, Manaus, AM, Brazil. 2-Programa de Pós-graduação em Biotecnologia e Recursos Naturais da Amazônia, UEA, 69050-010, Manaus, AM, Brazil

Endophytic fungi of the genus *Diaporthe* are prolific producers of bioactive metabolites, including sterols, terpenoids, and polyketides. In this context, this study aimed to investigate the antimicrobial potential of *D. hongkongensis* isolated from *Minquartia guianensis* (Coulaceae), focusing on the isolation and characterization of bioactive constituents. The fungus was reactivated on Sabouraud Dextrose Agar (30 °C, 7 days) and subjected to submerged fermentation in Erlenmeyer flasks totaling 6.5 L of Sabouraud Dextrose Broth with 0.2% yeast extract at 30 °C for 21 days under agitation (120 rpm). Post-fermentation biomass was extracted sequentially using dichloromethane (DCM), ethyl acetate (EtOAc), and methanol (MeOH). The resulting crude extracts were screened for antibacterial and antifungal activities against reference strains. The EtOAc and DCM extracts selectively inhibited *Staphylococcus aureus* (MIC 500–1000 µg/mL), while no significant activity was observed against *Escherichia coli* or *Pseudomonas aeruginosa*. The MeOH extract showed moderate antifungal activity against *Candida albicans* and *Cryptococcus gattii* (MIC 800 µg/mL). Guided by these results, the DCM–EtOAc extract was subjected to chromatographic separation, affording the isolation of ergosterol peroxide. The compound was identified by NMR spectroscopic analyses (¹H and ¹³C NMR, DEPT-135, COSY, HSQC, HMBC), confirming its structure as 5 α ,8 α -epidioxyergosta-6,22-dien-3 β -ol. The compound's distinctive 5 α ,8 α -peroxide bridge functions as a pharmacophore that mediates its antimicrobial activity by interacting with membrane sterols, destabilizing lipid bilayers, and inducing reactive oxygen species that disrupt microbial homeostasis. Isolated from fungi and plants, ergosterol peroxide exhibits broad-spectrum bioactivity, with antifungal and antitubercular activities among the most consistently documented. Its identification in *D. hongkongensis* is notable, marking the first report of this metabolite from the species and underscoring the ecological versatility and metabolic richness of the genus *Diaporthe*, while reinforcing evidence that endophytic strains from biodiverse ecosystems are valuable reservoirs of pharmacologically active sterols. The authors would like to thank CAPES, CNPq, FAPEAM, FINEP and LTQPN-INPA.

Keywords: sterol, endophytic fungi, submerged fermentation, bioprospecting, NMR

