Biophysical characterization of antibacterial compounds derived from pathogenic fungi Ganoderma boninense

ABSTRACT

There have been relatively few studies which support a link between Ganoderma boninense, a phytopathogenic fungus that is particularly cytotoxic and pathogenic to plant tissues and roots, and antimicrobial compounds. We previously observed that liquid-liquid extraction (LLE) using chloroformmethanol-water at a ratio (1:1:1) was superior at detecting antibacterial activities and significant quantities of antibacterial compounds. Herein, we demonstrate that antibacterial secondary metabolites are produced from G. boninense mycelia. Antibacterial compounds were monitored in concurrent biochemical and biophysical experiments. The combined methods included high performance thin-layer chromatography (HPTLC), gas chromatography-mass spectrometry (GC-MS), high-performance liquid chromatography (HPLC), fourier transform infrared (FTIR), and nuclear magnetic resonance (NMR) spectroscopy. The antibacterial compounds derived from mycelia with chloroform-methanol extraction through LLE were isolated via a gradient solvent elution system using HPTLC. The antibacterial activity of the isolated compounds was observed to be the most potent against Staphylococcus aureus ATCC 25923 and multidrug-resistant S. aureus NCTC 11939. GC-MS, HPLC, and FTIR analysis confirmed two antibacterial compounds, which were identified as 4,4,14α-trimethylcholestane (m/z = 414.75; lanostane, $C_{30}H_{54}$) and ergosta-5,7,22-trien-3β-ol (m/z = 396.65; ergosterol, $C_{28}H_{44}O$). With the aid of spectroscopic evaluations, ganoboninketal (m/z = 498.66, $C_{30}H_{42}O_6$), which belongs to the 3,4-seco-27-norlanostane triterpene family, was additionally characterized by 2D-NMR analysis. Despite the lack of antibacterial potential exhibited by lanostane; both ergosterol and ganoboninketal displayed significant antibacterial activities against bacterial pathogens. Results provide evidence for the existence of bioactive compounds in the mycelia of the relatively unexplored phytopathogenic G. boninense, together with a robust method for estimating the corresponding potent antibacterial secondary metabolites