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## Investigation of the antibacterial properties of the bracket fungus *Ganoderma lucidum*

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The wound healing properties of aboriginal medicinal plants is well established amongst native Australians. *Ganoderma lucidum*, a bracket fungus indigenous to Queensland's tropical rainforests, is also common to Japan (known as Red Reishi) and China (Lingzhi). Traditionally, *G. lucidum* was used to heal wounds and ensure smooth tissue regeneration. As such, we aim to evaluate the bactericidal properties of *G. lucidum* with regards to reducing microbial load in a chronic wound. Bioactive compounds were extracted separately with 90% v/v ethanol, absolute methanol and deionized (d.i.) water, submitted to separate protocols and obtained as lyophilized crude extracts (denoted as primary extracts). Next, the extracts were dissolved in d.i. water to various concentrations (10, 25, 50 mg/mL) and assessed for their antimicrobial activity against a range of common wound-colonizing bacteria in the well diffusion assay. All assays were performed in triplicate (n=3). Zones of inhibition were measured (mm) and expressed as  $\pm$ SEM. Positive controls: trimethoprim+sulfamethoxazole for MRSA, penicillin G for MSSA, gentamicin for *Escherichia coli*, *Pseudomonas aeruginosa* and *Klebsiella pneumoniae*, erythromycin for *Streptococcus pyogenes* and *Bacillus cereus*. After 24 hours and at a concentration of 50 mg/mL, in the well diffusion assays, all the Gram-positive bacteria resulted susceptible to the primary extracts with the exception of *S. pyogenes*. MRSA was most inhibited by the ethanol extract, which elicited an IZ of  $12.7\pm 0.3$  mm, by the hot water (IZ  $12.1\pm 0.7$  mm) and cold water extract (IZ  $11.4\pm 1.3$  mm), while the methanol extract was less effective (IZ  $8.3\pm 0.3$  mm). MSSA elicited from the methanol extract an IZ of  $12.0\pm 0.0$  mm, from the hot water extract an IZ  $11.3\pm 1.0$  mm and from the cold water extract an IZ  $10.8\pm 0.6$  mm, while caused a less pronounced IZ from the ethanol extract ( $8.7\pm 1.3$  mm). *B. cereus* stimulated a similar IZ from the ethanol, methanol and cold water extracts (respectively  $9.4\pm 0.7$  mm,  $9.8\pm 0.6$  mm,  $9.7\pm 0.3$  mm), while elicited a smaller IZ from hot water extract ( $6.5\pm 0.2$  mm). *S. pyogenes* prompted a greater IZ of  $15.7\pm 1.8$  mm from the cold water extract and a lesser IZ from the methanol extract ( $11.7\pm 0.5$  mm) and the hot water ( $10.5\pm 2.0$  mm). The water extracts were able to inhibit successfully the only Gram-negative bacterium *E. coli* with the cold water extract (IZ  $9.5\pm 0.5$  mm) performing better than the hot water extract ( $8.4\pm 0.3$  mm). The results clearly demonstrate that the primary extracts obtained from *G. lucidum* at a concentration of 50 mg/mL, elicit bactericidal activity against Gram positive and Gram negative bacteria.

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