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ANTIBIOFILM ACTIVITY OF *Acronychiapedunculata* AGAINST METHICILLIN-RESISTANT *Staphylococcus aureus*

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Acronychiapedunculata (L.)Miq. (Rutaceae) is reported to display antimicrobial activity against a range of microbes. However, the effect of the plant extracts against infectious biofilms has not been described. We report the results of preliminary studies on the anti-adhesion activity of *A. pedunculata* extracts on the formation of biofilms by a clinical isolate of methicillin-resistant *Staphylococcus aureus* (MRSA) and a standard strain of methicillin-sensitive *S.aureus* NCTC 6571.

Air-dried powdered leaves and stem-bark of *A. pedunculata* were extracted into methanol using a sonicator (30 °C for 30 min) or a Soxhlet apparatus (65 °C for 24 h). The extracts were tested for antibacterial activity against *S. aureus* and MRSA using the agar well diffusion assay (well diameter, 9 mm). Minimum inhibitory concentration (MIC) values of the extracts were determined by the broth dilution method. Anti-adhesion activity was measured as colony forming units (CFU) by growing biofilms on a catheter tube in the presence of extracts at MIC values and culturing the viable bacteria present in the dislodged biofilms to form colonies on a Muller-Hinton agar plate.

The plant displayed large zones of inhibition (*S. aureus* and MRSA) at relatively low concentrations of leaf (1.0 mg/ml) and stem-bark (0.7 mg/ml) extracts: 14-16 mm (sonicator extracts) and 17-18 mm (Soxhlet extracts). The MIC values were consistently lower for the Soxhlet extracts (2-64 ppm) than for the corresponding sonicator extracts (16-128 ppm). The MIC values of the stem-bark extracts (2-64 ppm) were lower than the corresponding values of the leaf extracts (64-128 ppm). The lowest MIC values were observed for the Soxhlet stem-bark extracts (4 ppm, *S. aureus*; 2 ppm, MRSA), and the highest MIC values were for the sonicator leaf extracts (64 ppm, *S. aureus*; 128 ppm, MRSA). The results show that the stem-bark extract prepared using the Soxhlet apparatus had the highest antibacterial activity.

In the antibiofilm assay with MRSA, the CFU values were 0.7×10^3 and 1.2×10^3 ml⁻¹ in the presence of Soxhlet stem-bark extract (at 2 ppm) and sonicator stem-bark extract (at 64 ppm), respectively; the CFU values for the negative control (nutrient broth medium) and positive control (vancomycin, 1 ppm) were 1.8×10^3 and 1.0×10^3 ml⁻¹, respectively. There was a reduction in the adherence of MRSA on the catheter tube at the MIC values of all the extracts. Further, the extracts showed higher anti-adhesion activity towards MRSA than *S. aureus*. However, increasing the concentration of the extracts did not always improve the antibiofilm activity.