

Antimicrobial activity of skin secretions of selected frog species in Sri Lanka against fungal and bacterial pathogens of humans

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Introduction and Objectives: Amphibians have immune systems that include both innate and adaptive immunity. Their defense mechanisms involve the production of various molecules and secretions including antimicrobial peptides (AMPs). AMPs have gained attention as a good source of novel antimicrobial agents. The present study aimed to (i) isolate skin secretions from selected frog species in Sri Lanka and test their antimicrobial activity against representatives of Gram-positive and Gram-negative bacterial strains and fungal species, and (ii) determine the minimum inhibitory concentrations (MICs) of frog skin secretions against selected bacteria and fungi.

Methods: Skin secretions of the frog species *Duttaphrynus melanostictus*, *Minervarya agricola*, *Microhyla mihintale*, *Polypedates maculatus* and *Hydrophylax gracilis* collected from Kaludiya pokuna, Mihintale were investigated for antimicrobial activity for the first time in Sri Lanka. Amphibian skin secretions were collected using a non-lethal method with chemical stimulation using norepinephrine. Antimicrobial activity was evaluated using the agar well diffusion method, broth method and broth macro dilution method against *Staphylococcus aureus* and Methicillin-resistant *Staphylococcus aureus* (MRSA), *Escherichia coli* and *Candida albicans*. The secretions collected were concentrated by freeze-drying and re-dissolved in 2 ml of sterile distilled water to determine the Minimum Inhibitory Concentration (MIC). The test was conducted in triplicate.

Results: Skin secretions of *H. gracilis* exhibited antimicrobial activity against clinical isolates of *S. aureus* and Methicillin-resistant *S. aureus* (MRSA) in the broth method. The minimum inhibitory concentration was 50% of original extract for *S. aureus* and inhibition was not obtained for *E. coli*. Faintly noticeable inhibitions were observed for *C. albicans*.

Conclusions: Skin secretions of *H. gracilis* exhibited antimicrobial activity against *S. aureus* and MRSA. These findings suggest that the active skin secretions of Sri Lankan frogs can be promising sources of animal-derived antimicrobial agents.

Keywords: Amphibian, Antimicrobial peptides, *Hydrophylax gracilis*, MRSA

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