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Isolation, Screening and Identification of Frog Cutaneous Bacteria for Anti-Batrachochytrium dendrobatidis Activity

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Abstract : Mitigating strategies using symbiotic cutaneous bacteria is one of the major concerns in the conservation of amphibian population. Batrachochytrium dendrobatidis is the causative agent of chytridiomycosis associated with mass mortality and amphibian extinctions worldwide. In the Philippines, there is a lack of study on the cutaneous bacteria of Philippine amphibians that may have beneficial effects to ward off the deadly fungal infection. In this study, cutaneous bacteria from frogs were isolated and examined for anti-B. dendrobatidis activity. Eight species of frogs were collected at Mt. Palay-palay Mataas na Gulod National Park in Cavite, a site positive for the presence of B. dendrobatidis. Bacteria were isolated from the skin of frogs by swabbing the surfaces of the body and inoculated in Reasoner's 2A (R2A) agar. Isolated bacteria were tested for potential inhibitory properties against B. dendrobatidis through zoospore inhibition assay. Results showed that frog cutaneous bacteria significantly inhibited the growth of B. dendrobatidis in vitro. By means of 16S rRNA gene primers, the anti-B. dendrobatidis bacteria were identified to be Enterobacter sp., Alcaligenes faecalis and Pseudomonas sp. Cutaneous bacteria namely Enterobacter sp. (isolates PLd33 and PCv4) and Pseudomonas (isolate PLd31) remarkably cleared the growth of B. dendrobatidis zoospore in 1% tryptone agar. Therefore, frog cutaneous bacteria inhibited B. dendrobatidis in vitro and could possibly contribute to the immunity and defense of frogs against the lethal chytridiomycosis.

Keywords: Batrachochytrium dendrobatidis, cutaneous bacteria, frogs, zoospore inhibition assay

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