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Probiotic Potential of Actinomycetes from Shrimp Gut and Shrimp-Farming Pond Sediments

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Background: The use of probiotics for improving water quality and enhancing productivity is an emerging eco-friendly strategy in shrimp farming. Heterotrophic bacteria inhabit shrimp-farming-pond sediments, and the shrimp gut exhibit remarkable probiotic potential through the production of extracellular enzymes involve in bioremediation of organic waste and digestion of complex nutrients in feed. Actinomycetes is a group of heterotrophic bacteria reported to have strong extracellular enzyme-producing capabilities. However, strains associated with shrimp-farming ponds and the shrimp gut remain underexplored in Sri Lanka.

Objective: This study aimed to evaluate the probiotic potential of Actinomycetes isolated from the gut of cultured shrimps (*Litopenaeus vannamei* and *Penaeus monodon*) and pond sediments in terms of their extracellular enzyme production.

Methods: Samples were collected from shrimp-farming ponds in Mundalama, Sri Lanka. Intestinal segments of shrimps were obtained aseptically. Both intestinal and sediment samples were heat-treated (50 °C, 60 min) to eliminate less heat-resistant non-Actinomycetes. Then, CaCO₃ was added into the samples for enriching Actinomycetes. After the enrichment, isolation was carried out using the serial dilution method on starch casein agar supplemented with selective agents, cycloheximide (80 µg/ml) and nalidixic acid (75 µg/ml). Colonies exhibiting characteristic morphology of Actinomycetes were purified and confirmed with biochemical tests. All isolates were screened for extracellular enzymes—protease, amylase, cellulase, and lipase—on the formation of a halo zone on skim milk, starch, carboxymethyl cellulose, and phenol red–olive oil agars, respectively, following spot inoculation. Enzymatic activity was quantified as the Enzymatic Index (EI), the ratio of halo zone to colony diameter.

Results: Nineteen isolates were obtained (fourteen sediment-derived and five gut-derived), of which ~80% exhibited notable enzymatic activity (EI ≥ 2.0) for at least two enzymes. Cellulase activity was detected in 68% of the isolates, with the maximum EI of 8.2. Amylase and protease activities were observed in 73% of the isolates, with maximum EI of 5.1 and 3.8, respectively. Although lipase activity was the most prevalent (89%), its maximum EI (2.7) was lower than the lowest EI of other enzymes tested. All the gut-derived isolates exhibited four of the enzyme activities.

Conclusion: Strong extracellular enzymatic activity exhibited by Actinomycetes from shrimp-farming ponds and the gut of cultured shrimp suggests their high probiotic potential and suitability for inclusion in shrimp feed formulae.

Keywords: Actinomycetes, Extracellular enzymes, Shrimp farming