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Occurrence of lipase activity in the pitcher juice of *Nepenthes distillatoria* (bandura)

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Nepenthes distillatoria (Family Nepenthaceae), an insectivorous pitcher plant endemic to Sri Lanka, is believed to contain a mixture of hydrolytic enzymes in its pitcher. These enzymes are believed to be primarily responsible for efficient digestion of insect carcasses inside the pitcher fluid. In this study we partially characterized lipase activity detected in the crude pitcher fluid of *N. distillatoria*.

Crude juice from pitchers of *N. distillatoria* was collected from Hakurugala forest patch at Ruwanwella. A simple colorimetric method for characterizing lipase in the pitcher fluid of *N. distillatoria* was developed, using 2,3-dimercapto-1-propanol tributyrates (DMPTB) as the substrate and 5,5'-dithiobis (2-nitrobenzoic acid) (DTNB) as the chromogenic reagent. Optimum pH and optimum temperature for lipase activity were determined by incubating the reaction mixture at different pHs and at different temperatures. Crude pitcher fluid was incubated in buffers at different pHs (from pH 2.0 -10.0) over a period of two weeks at 4°C to determine pH stability. To determine the thermal stability, the crude enzyme mixture was incubated in pH 4.0 buffer at different temperatures for a period of two weeks. Lipases in the crude pitcher fluid were partially purified using a DEAE cellulose column.\

A detectable lipase activity was observed in the crude *Nepenthes* pitcher juice. The crude pitcher fluid had two pH optima for lipase activity at pH 4.0 and pH 5.0 indicating the presence of more than one lipase. The optimum reaction temperature was in-between 35°C - 40°C. Within the two week period tested, the lipase activity at 4°C and room temperature was not significantly reduced. In contrast, the percentage remaining activity after two weeks at 37°C and beyond was significantly low. At pH 5.0 and pH 6.0, the lipase activity was significantly stable. Enzyme activity was lost within 4 days at pHs 2, 3, 8, 9, and 10. DEAE cellulose chromatography resulted in two sharp peaks at NaCl concentrations 0.12 M and 0.18 M, indicating the presence of at least two lipases in the crude juice. Presence of these enzymes probably has a significant role in the digestion of complex organic matter in insect bodies into simpler absorbable molecules. Further studies are in progress on purification and characterization of the individual enzymes.

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