Endolichenic Fungi in *Usnea* sp. from Hakgala Montane Forest in Sri Lanka as a Novel Source of Bioactive Natural Products

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Abstract

Endolichenic fungal strains were isolated from the abundant lichen, *Usnea* in Hakgala tropical montane forest, using surface sterilization method. Eighteen fungal strains have been isolated from *Usnea* (Curvularia sp., Fusarium sp., Nigrospora sp., Cladosporium sp., Chrysosporium sp. 1, Chrysosporium sp. 2, Broomella sp., Aspergillus sp.1 and sp. 2., Acremonium sp. and eight dark sterile sp.). Substrate utilization patterns, fungicidal effects and insecticidal effects of these fungi were evaluated and the results revealed that all the test fungi were able to produce at least one enzyme to utilize the test substrates. The highest fungicidal effect was observed from *Fusarium* sp. and the highest insecticidal effect was shown by *Curvularia* sp.

Introduction

Interest in fungal endophytes as potential producers of novel and biologically active products has increased in the last decade and a recent comprehensive study indicated that 51% of the biologically active substances isolated from fungal metabolites were previously unknown. Many new bioactive natural products have been found from fungal metabolites, including compounds with antimicrobial, anticancer and insecticidal activities (Li et al, 2005). Endolichenic fungi are living within the asymptomatic lichen thallus much the same way as endophytic fungi live within healthy plant tissues. The potential of endolichenic fungi as novel sources of bioactive secondary metabolites is still largely unexplored. Many studies have been conducted regarding bioactivities of secondary metabolites of plant endophytes and only few studies were carried out to investigate the bioactivity studies of endolichenic fungi.

Methodology

Lichen *Usnea* from Hakgala montane forest was collected randomly in previously demarcated five study plots. Collected lichens were packed in sterile polythene bags and transported to University of Kelaniya. Lichen species were cleaned using tap water and surface sterilized by consecutive immersion for 10 seconds in 95% ethanol, 3 minutes in 0.5% sodium hypochlorite and 30 seconds in 75% ethanol. The thalli were surface dried with sterile filter papers and cut aseptically into small segments. Lichen particles of 1 mm were then plated (20 pieces from each of the 5 replicate samples) on 2% Malt Extract Agar (MEA) supplemented with 0.01% streptomycin. The plates were incubated at room temperature (30 °C) for 7-10 days. Fungi growing from each lichen particle were isolated into pure cultures. Slides