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Extracellular extracts of antagonistic fungi,*Trichoderma longibrachiatum* and *Trichoderma viride*, as larvicides against dengue vectors, *Aedes aegypti* and *Aedes albopictus*

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Abstract

Aedes aegypti and *Aedes albopictus* are the responsible vectors of transmitting dengue in Sri Lanka. Excessive use of chemical pesticides causes insecticide resistance.Therefore, mosquito biolarvicides remain to be an important method for mosquito control. The use of fungal metabolites can be considered a tool to overcome the issues related to insecticide resistance and environmental pollution. The present study focused on the evaluation of the mosquito larvicidal efficacy of *Trichoderma longibrachiatum* and *Trichoderma viride* aqueous and crude extracellular metabolites against *Ae.aegypti* and *Ae.albopictus* under the laboratory conditions. Fungi were grown in Richards' broth medium for collection of fungal biomass. Eight test concentrations of extracellular aqueous and crude fungal filtrates in a range from 12.5 gL−1 to 175 gL−1 were prepared and batches of 25 laboratory-reared third instar larvae of *Ae.aegypti* and *Ae.albopictus* were exposed to each test concentration separately with three replicates. Control bioassays were conducted with distilled water and larval mortality was recorded after 24 hours and 48 hours of exposure periods.

The study revealed that LC_{50} values and LC_{90} values for extracellular crude metabolites for two fungal species were lower than the values obtained for extracellular aqueous extracts. All the LC_{50} values obtained for *Ae.albopictus* were lower than that of A*e.aegypti*. LC_{50} values obtained for *T.viride* aqueous extracellular metabolites were 81.46 gL^{-1} and 87.75 gL^{-1}in 24 hours, 70.66 gL^{-1} and 77.93 gL^{-1} in 48 hours for *Ae. albopictus* and *Ae. aegypti* respectively. LC_{50} values obtained for *T.longibrachiatum* aqueous extracellular metabolites were recorded as 103.35gL^{-1} and 108.79gL^{-1} in 24 hours and 93.05gL^{-1} and 102.1gL^{-1} in 48 hours for *Ae.albopictus* and *Ae.aegypti* respectively. It was revealed that LC_{50} values obtained for *T.viride* crude extracellular metabolites were $34.42gL^{-1}$ and $47.27gL^{-1}$ in 24 hours, $30.29gL^{-1}$ and $41.56gL^{-1}$ in hours for *Ae.albopictus* and *Ae.aegypti* respectively. LC_{50} values obtained for *T.longibrachiatum* crude extracellular metabolites were $49.84gL^{-1}$ and $59.24gL^{-1}$ in 24 hours, $40.73gL^{-1}$ and $51.49gL^{-1}$ in 48 hours for *Ae.albopictus* and *Ae.aegypti* respectively. It can be concluded that there is a potential for using *T.viride* as an effective larvicide against dengue mosquitoes.

Introduction

Mosquito-borne diseases become a greater issue among all other diseases in public health. Millions of people suffer from many mosquito-borne diseases such as dengue, malaria, chikungunya, zika, lymphatic filariasis, and Japanese encephalitis. With increasing urbanization, land use patterns, unplanned garbage disposal, unplanned irrigation, sewage disposal, and changing environmental factors make a favorable environment for the colonization of mosquitoes very rapidly which leads to the major outbreaks of many mosquito-borne diseases (Ortiz *etal.*, 2021). Among them dengue and dengue haemorrhage fever play major public health problems in Sri Lanka, being endemic within the country since 1960s (Sirisenaand Noordeen,2013). The established vectors in the country are *Ae.aegypti* and *Ae.albopictus* which are belonged to the subgenus *Stegomyia*, carrying four subtypes of the dengue virus (Lambrechts *etal.,* 2010).

The controlling agenda of dengue vector mosquitoes is timely challenging due to very fast spread, development of resistance against synthetic insecticides, adverse effects on non-target populations particularly on mammals, non-biodegradable nature and environmental pollution (Govindarajan *etal.,* 2016; Anupam *etal.,* 2012). Strategies that rely on their control; use of synthetic herbal and microbial larvicides, adult repellents, Sterile Insect Technique (SIT), bacterial symbiont-based techniques, and transgenic mosquitoes are not fully successful (Suresh *etal.,*2022). The solution for these arising problems relies on searching for ecofriendly and effective alternatives to control dengue vectors.

Fungi have excellent potential for the production of mycopesticides due to their safety, relatively limited host range, and their ability to secrete bioactive secondary metabolites (Govindarajan *etal.,* 2005). Extracellular secondary metabolites of many fungi have been screened as a larvicide for many mosquito species (Vijayanand Balaraman,1991). *Trichoderma* spp. are soil-borne, spore-forming, filamentous Ascomycetes with vast economic importance due to their production of industrial enzymes, secondary metabolites and their ability to act as biological control agents (Vinale *etal.,*2006; Hamdanand Jasim,2021). Current study was conducted to evaluate the potential use of extracellular metabolites of *Trichoderma longibrachiatum* and *Trichoderma viride* as novel vector controlling strategies against third instar larvae of *Ae.aegypti* and *Ae.albopictus.*

Section snippets

Maintain laboratory cultures of Trichoderma longibrachiatum and Trichoderma viride