## **RESEARCH ARTICLE**

## Characterisation of entomopathogenic fungi used in the biological control programme of *Diaphorina citri* in Mexico

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The production of biological insecticides requires the identification and selection of candidate isolates of entomopathogenic fungi (EPF). The objective of this study was to characterise by virulence factors, single-spore strains of the CHE-CNRCB 303, 305, 307 strains initially identified as *Isaria fumosorosea* and the 224 strain of *Metarhizium anisopliae* to identify those with outstanding quality parameters that can serve to improve the control programme for *Diaphorina citri* (Hemiptera: Liviidae) in Mexico. The size of conidia and phialides was evaluated. The internal transcribed spacers established molecular identity. The selection of single-spore strains was done using radial growth (RG), the production of conidia (PC) and germination (G) rates. Principal component analysis (PCA) was used to determine the correlation between the variables and as screening criterion. Molecular and morphometric analysis confirmed that isolates 303, 305 and 307, and their singlespore strains correspond to the Isaria javanica species. The 224 strain and its single-spore strain were identified as M. anisopliae sensu lato. The statistical analysis of the RG, PC and G variables showed significant differences ( $P \le 0.05$ ) between single-spore strains and their multi-spore strains. PCA showed a correlation between the RG and PC. The assay with Pr1 proteases associated with the surface of the conidia showed significant differences ( $P \le 0.05$ ) for singlespore strains of *I. javanica* 303/2, 305/1 and 305/5 compared to the original strains. The characterisation of genetically homogeneous populations of EPF will allow us to obtain improved isolates, which are needed for the mass production of mycoinsecticides.

Keywords: single-spore strains; virulence factors; Isaria fumosorosea; Isaria javanica; Metarhizium anisopliae; Diaphorina citri

## 1. Introduction

The Servicio Nacional de Sanidad, Inocuidad y Calidad Agroalimentaria (SENA-SICA), through the Dirección General de Sanidad Vegetal (DGSV) of the Government of Mexico, developed a biological control programme for the management of the Asian citrus psyllid (ACP) [*Diaphorina citri* Kuwayama (Hemiptera: Liviidae)] through the use of entomopathogenic fungi (EPF) in commercial orchards. *D. citri* is the vector of the bacteria *Candidatus liberibacter* spp., which cause Huanglongbing disease (HLB), commonly known as citrus

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