Título: Genetic transformation of *Fusarium moniliforme* via *Agrobacterium tumefaciens*

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This work describes for the first time an efficient and successful methodology for genetic transformation via *Agrobacterium tumefaciens* of the endophytic fungus *Fusarium moniliforme*. This fungus was isolated in high frequency from sugarcane plants. Sugarcane is a very important culture, especially in Brazil, that has historical, economic, social and political relevance, being the main source of raw material to produce sugar and alcohol. Many fungal species live associated to plants, either on the surface, within tissue and organs or in the rhizosphere, establishing interactions that can vary from mutualism to parasitism. By definition, endophytic fungi live inside the plant and do not cause harm to them. The study of microbial communities associated to sugarcane may unravel unknown functional roles boosted by this interaction. Mycelia of *F. moniliforme* were transformed via *A. tumefaciens*, which carried the plasmid pCAMDsRed, containing both genes encoding for hygromicin resistance (*hph*) as well as *DsRed.* The optimization of the agro-transformation protocol was performed testing two different conditions: inductor agent acetoseringone concentration (AS) and co-culture time. Results demonstrated that the best condition occurred with the utilization of Millipore Nylon Hybond membrane, 400 $\mu M.mL^{-1}$ of AS and 48 hours as co-culture time. The generated transformants expressed the gene encoding for the *DsRed*, a red fluorescent protein used as molecular reporter. PCR technique confirmed the *hph* gene insertion into the *F. moniliforme* genome. In addition, the presence of *DsRed* inside the mycelia was confirmed by epifluorescence optical microscopy and one of the transgenic fungus showed higher amilolitic activity compared with the wild type, according to enzymatic analysis. The establishment of a transformation method for *F. moniliforme* opens a range of possibilities and facilitates to the study of insertional mutagenesis and genetic knockouts, in order to identify key genes involved in the plant-endophyte interaction.

Palavras-chave: *Fusarium moniliforme*, Genetic transformation, *Agrobacterium tumefaciens*, Endophytes, Sugarcane

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