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PCR-based assay for species-specific detection of *Fusarium solani* f. sp. *piperis* from soil and roots of black pepper (*Piper nigrum*) / ENSAIO BASEADO EM PCR PARA DETECÇÃO ESPECÍFICA DE *Fusarium solani* f. sp. *piperis* A PARTIR DE SOLO E RAÍZES DE PIMENTA DO REINO (*Piper nigrum*). S. S. COSTA¹; G. M. MOREIRA²; L. H. PFENNING¹.
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The main disease of black pepper (*Piper nigrum*) in Brazil is caused by *Fusarium solani* f. sp. *piperis* (Fpip) a member of the *Fusarium solani* species complex - FSSC. The disease diagnosis is problematic because different phylogenetic lineages of FSSC are associated with black pepper, what preclude the pathogen be identified based solely on morphology. We developed a specific primer pair to detect and identify Fpip by PCR, and to test its sensitivity and PCR protocols for the ability to detect the pathogen. Specific primers, Fpiper1F/Fpiper1R, were synthesized based on interspecific variations in the EF1- α gene sequences of species in the FSSC. Primer specificity was tested using conventional PCR for 18 isolates of Fpip, 24 representatives of other lineages of the FSSC associated with black pepper, and eight strains of other *Fusarium* species. The sensitivity of primers was evaluated using two methods: amplification of genomic DNA diluted in nine steps down 1 fg, and DNA extracted from 1 g of soil inoculated with 100 μ L of macroconidia suspension diluted at 10^1 to 10^7 . Conventional and nested PCR, in which the primers EF1/EF2 were used for the first amplification and specific primers for the second one, were used. Specific primers were also tested with extracted DNA from naturally infected soil and black pepper root samples using nested PCR approach. Primers amplified only a single PCR band of 230 bp from Fpip in both specificity test and from naturally infected soil and roots. In sensitivity test, nested PCR was more sensitive than conventional PCR. Detection sensitivity was 1 fg of purified target DNA template and soil infected with 10^1 macroconidia/g. The specific primers will facilitate the detection and identification of Fpip in propagative material as well as in field-grown black pepper and soil. Financial Support: FAPEMIG and CAPES.

Key words: Diagnosis; *Fusarium solani* species complex; Nested PCR; Specific primers.