ITS-based detection and quantification of Alternaria spp. in raw and processed vegetables by real-time quantitative PCR - DTU Orbit (26/01/2019)

A TaqMan real-time polymerase chain reaction (PCR) method was developed for specific detection of Alternaria spp. in foodstuffs. The method uses Alternaria-specific primers and probe targeting the internal transcribed spacer regions ITS1 and ITS2 of the rRNA gene, and a positive amplification control based on 18S rRNA gene. The applicability of the real-time PCR protocol was assessed through analysis of 190 commercial food samples, including 80 fresh fruit and vegetable samples and 110 processed foodstuffs. The assay demonstrated the presence of Alternaria spp. DNA in 46 out of the 80 raw samples (57.5%) and in 66 out of the 110 processed samples (60%), enabling quantitative detection of Alternaria spp. DNA at levels as low as 1 CFU/g. The estimated Alternaria counts obtained by real-time PCR showed a good relationship (R2 = 0.9006, P <0.01) with the Alternaria counts obtained by plating on Potato Carrot Agar (PCA). The developed real-time PCR assay provides a useful tool for early detection of Alternaria spp. and could be applied as a quality and biosecurity marker of raw materials and final products in the fruits and vegetables processing industries.

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