Title Sooty blotch and flyspeck of apple: Assessment of an RFLP-based identification technique and adaptation of a warning system for the Upper Midwest

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Abstract

Sooty blotch and flyspeck (SBFS) of apple, a disease caused by more than 30 species of fungi, reduces crop value by blemishing the fruit surface. This study investigated two research tools designed to improve identification of SBFS fungi and management of the disease.

The first objective was to validate a PCR-based method to identify SBFS pathogens. Members of the sooty blotch and flyspeck (SBFS) disease complex are challenging to identify by traditional mycological methods that rely on agar-plate isolation and morphological description. Identification using a PCR-RFLP assay was investigated as an alternative to culturing. The method involved amplification of the internal transcribed spacer region of ribosomal DNA using a Capnodiales order-specific reverse primer paired with a universal forward primer, followed by digestion using the *Hae* III restriction enzyme. When applied to 24 SBFS species from a survey in the Midwest U.S., the PCR-RFLP assay produced 14 unique band patterns, all specific to genus. The technique also identified SBFS fungi from DNA extracted directly from colonies on apples. The PCR-RFLP assay streamlined the identification process by circumventing the requirement for culturing, and should be a valuable tool for further ecological studies of the SBFS disease complex.

The second objective was to adapt a SBFS warning system for the Upper Midwest. The Sutton-Hartman warning system, developed in the Southeast U.S., uses cumulative hours of leaf wetness duration (LWD) to predict the timing of the first appearance of SBFS signs. In the Upper Midwest, however, this warning system experienced sporadic control failures. To determine if other weather variables were useful predictors of SBFS appearance, hourly LWD, rainfall, relative humidity (RH), and temperature data were collected from orchards in IA, WI and NC. Timing of the first appearance of SBFS was determined by scouting weekly for disease signs. Receiver operating characteristic curve analysis revealed that cumulative hours of RH$\geq$97% was a more conservative and accurate predictor than cumulative LWD for the Upper Midwest. The results suggest that the performance of the SBFS warning system in the Upper Midwest could be improved if cumulative hours of RH$\geq$97% were substituted for cumulative hours of LWD to predict the first appearance of SBFS.