Diseases Caused by Fungi and Fungus-Like Organisms

First Report of Atypical Scab Caused by *Venturia asperata* on Apple in China

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Funding: This work is supported by the fund for China Agriculture Research System (No. CARS-30). Plant Dis. 105:1858, 2021; published online as <https://doi.org/10.1094/PDIS-11-20-2431-PDN>. Accepted for publication 8 January 2021.

Apple cultivar ‘Huangtaiping’ (*Malus pumila* Mill.) is grown widely in northern China for the production of jellies, preserves, and cider. In 2018, atypical scab symptoms were observed on fruits of Huangtaiping in Heilongjiang Province of China. The disease incidence was estimated at approximately 0.4%. Symptoms were scab-like black spots (3 to 5 mm diameter) distinct from scab caused by *Venturia inaequalis*. Conidia were generally produced on lesions, and using a modified microscope (Goh 1999), a single spore was picked up from each sample on a water agar plate with a glass needle and then transferred to potato dextrose agar (PDA) amended with lactic acid (0.50 ml/liter) and sulfate streptomycin (0.20 g/liter). Fifteen isolates were obtained and incubated at 21°C for 6 weeks in darkness on PDA. The colonies on PDA were gray-black with circular morphology and floccose texture, which were similar with the characteristics of *Venturia asperata* described previously (Turan et al. 2019). The conidia were cylindrical to fusiform, zero to one septate, yellowish and $19.7 (13.5 \text{ to } 25.8) \times 5.7 (3.6 \text{ to } 6.9) \mu\text{m}$ ($n = 10$) in size, which were larger than previously described ones (Turan et al. 2019). DNA of three randomly selected isolates was extracted by a modified SDS method (Ping et al. 2004). The internal transcribed spacer (ITS) region of rDNA of the three selected isolates was amplified with the primers ITS4/ITS5 (White et al. 1990), sequenced, and deposited in GenBank (MN958665, MN958666, and MN958667). BLAST analysis showed that the amplified sequences were identical and had 99.3% sequence identity with *V. asperata* (AF333447, MT459450, and MT459451), 95.4% sequence identity with *V.*

cerasi (MK810963 and MK810964), and 94.3% sequence identity with *V. carpophila* (MN958609, MN958610, and MN958611). In addition, the complete large subunit ribosomal RNA gene (*LSU*) was amplified with the primers LROR/LR5 (Vilgalys and Hester 1990), sequenced, and deposited in GenBank (MT845787, MT845788, and MT845789). BLAST analysis showed that the amplified sequences were identical and had 99.7% sequence identity with *V. asperata* (EF114711), 99.2% identity with *V. carpophila* (MT772296, MT845732, and MT845733), and 98% identity with *V. cerasi* (MK810848 and MK810849). Phylogenetic analysis based on concatenated ITS and *LSU* sequences showed that the tested isolates grouped with *V. asperata* strain 2349 in the same clade, and the closest species with *V. asperata* was *V. carpophila*, followed by *V. cerasi*. In July 2019, pathogenicity of the isolate VAHLJ3-1-1 was evaluated on Huangtaiping. A conidial suspension with a concentration of 5×10^5 conidia/ml was sprayed evenly on the surface of six fruits. To maintain high humidity, inoculated fruits were wrapped with a plastic bag (a cotton ball with water was placed in the plastic bag) to maintain wetness for 3 days. Six fruits sprayed with water were used as a control. Four weeks after inoculation, a similar symptom of atypical scab was observed on fruits of Huangtaiping, and *V. asperata* was isolated again from six inoculated fruits with reisolation frequency of 100% by the single-spore isolation, whereas no symptoms were observed on the control fruits. Based on the morphological and molecular identifications, the causal agent of atypical scab on Huangtaiping was identified as *V. asperata*. Apple scab is usually caused by *V. inaequalis* (Shen et al. 2020). However, apple scab has also been caused by *V. asperata* in Italy and France (Caffier et al. 2012; Turan et al. 2019). To the best of our knowledge, this is the first report of *V. asperata* associated with apple scab-like lesions in China. This information augments our knowledge of the spectrum of *Venturia* species associated with disease on apple fruit and will be a valuable foundation underpinning management strategies for this cultivar.

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The author(s) declare no conflict of interest.

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Keywords: apple, *Malus pumila*, atypical scab, *Venturia asperata*

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