First Report of *Alternaria* Black Spot Disease Caused by *Alternaria alternata* on the Invasive Weed *Solanum rostratum* in Xinjiang, China

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Solanum rostratum is a noxious weed, native to Mexico and the USA, which has invaded Liaoning, Jilin, Hebei, Inner Mongolia, Shanxi, Xinjiang and Beijing, China (Eminniya et al., 2013). In August 2015, foliar symptoms of yellowish to black spots were observed on plants of S. rostratum nearby an agricultural plantation in Changii, Xinjiang. The following year, about 17% of the 206 plants surveyed on about 0.2 ha of deserted farmland were infected from July-September (at 19-35°C under 29-97% RH). Symptoms of the disease appeared on about 70% of leaves on all the infected plants. Initially, some yellowish spots were observed on the abaxial dorsal leaves, which subsequently turned white and some dark spots appeared on the front and dorsal sections of the infected leaves; the spots gradually joined to some irregular larger spots, finally entire leaves fell or withered. After heavy rainfall, the adaxial region of some infected leaves, flowers and stems were observed to produce some dark spots. To isolate the pathogen, infected leaf tissues were surface-sterilized with 5% sodium hypochlorite for 3 min and rinsed with five changes of sterilized water, then transferred to potato dextrose agar (PDA) medium and cultured three to five days at 25°C, in the dark. In addition, single fungal spores were picked from infected tissues using a stereo microscope, and inoculated onto PDA. From which, three strains identical to the colony and conidial characteristics were isolated. The fungal colonies inocubated for 5 days were 8-9 cm in diameter, grey at the centre and pale at the margins. The conidia were brown, obclavate, obpyriform or ovoid to ellipsoidal, with 1-6 transverse septa and 0-3 longitudinal or obligue septa (14-42 × 6-12 µm; n=50). Three conidial suspensions (1 × 10⁶ conidia/ml) were prepared by harvesting conidia from three 2-week-old cultures grown in the dark at 25°C. Then, pathogenicity of the isolates was tested on single leaves of one month-old healthy seedlings. A 30µl drop of conidial suspension of each isolate was placed on each leaf of a pot of 50 one-month-old healthy seedlings in a greenhouse at 20-25°C and <50% RH. For the control treatment, 30 µl of sterilized water was placed on each leaf of a pot of 38 one-month-old healthy seedlings. Symptoms similar to naturally infected leaves developed on some leaves of 9 different weaker seedlings than others after 14 days of inoculation, then dark spots were produced on both sides of leaves flowers, stems and the fruits of all the 50 infected seedlings after 30 days of inoculation. Finally, all the seedlings withered. All control seedlings were asymptomatic. The fungus identical to the original isolates was re-isolated from the inoculated leaves fulfilling Koch's postulates. To further identify the fungus, a single-spore culture of one representative strain SRI S-1 was performed by PCR and sequencing using the internal transcribed spacer (ITS), glyceraldehyde 3-phosphate dehydrogenase (gpd) gene, Alternaria allergen a 1 (Alt a 1) and 28S large subunit ribosomal RNA (LSU) gene, as described previously (Hong et al., 2005; Paul et al., 2015). BLASTn analysis of the ITS (KX894536), gpd gene (KX894538), Alt a 1 gene (KX894537) and LSU (KX894539) obtained with sequences available in the GenBank database revealed 100% sequence identity, respectively, to Alternaria alternata (MF356593 for ITS, KR051396 for qpd, AY563301 for Alt a 1, and KP940477 for LSU). Therefore, based on both morphological and molecular identification, the strain SRLS-1 was identified as A. alternata. Similarly to that causes leaf spot of S. muricatum in Gansu, north-west China (Li et al., 2016). To our knowledge, this is the first report of A. alternata causing Alternatia black spot disease of S rostratum in China or elsewhere. The significance of our results is that this pathogen could be used as biocontrol agent against this invasive weed both in China and other countries, whereas the possibility of using this pathogen as a biocontrol agent should be validated experimentally.

References:

Eminniya, A. et al. 2013. Weed Sci, 61: 557. Hong, S. G. et al. 2005. Fungal Genet Biol, 42: 119. Paul, N. C. et al. 2015. Mycobiol, 43: 384. Li, J. B. et al. 2016. Plant Dis. 100(1): 224.



Fig.1 Diseased leaves of Solanum rustratum in the field in Changji, Xinjiang, China.

207x190mm (96 x 96 DPI)