Diseases Caused by Oomycetes

First Report on the Occurrence of an Aggressive Pathotype, 734, of *Plasmopara halstedii* Causing Sunflower Downy Mildew in Hungary

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Downy mildew of sunflower (*Helianthus annuus* L.) is caused by *Plasmopara halstedii* (Farr.) Berl. et de Toni, leading to significant losses in crop production worldwide. The number of new and more aggressive pathotypes has increased rapidly over the last 10 years in Europe (Bán et al. 2018; Virányi et al. 2015); therefore, constantly monitoring the distribution of races is an important task. As part of regular surveys in June 2019, severe downy mildew was identified in some regions, appearing as chlorotic lesions along the veins of the adaxial side and white sporulation on the abaxial side of the leaves of severely stunted hybrids containing *PI₆* and *PI₇* resistance genes. The identification of the pathogen was performed microscopically based on morphological characteristics (average size of sporangia: 28 × 20 µm). Disease incidence (the ratio of diseased plants) ranged between 10 and 30% per field in three regions and resulted in moderate yield loss. Isolates (defined as a lesion per leaf) were collected from four to eight infected leaves of each hybrid by region and stored at -70°C. Two, one, and one isolates of *P. halstedii* were selected and characterized from the southeastern (Békés County), northern (Nógrád County), and northeastern (Borsod-Abaúj-Zemplén County) regions of Hungary, respectively. The pathotype of the four isolates was determined using the international standardized nomenclature method reviewed by Trojanová et al. (2017), including nine sunflower differential inbred lines (HA-304, RHA-265, RHA-274, PMI-3, PM-17, 803-1, HAR-4, QHP2, and HA-335). Zoospores from frozen sunflower leaves were washed off into

bidistilled water, and the concentration was adjusted to 3.5 × 10⁴ sporangia/ml using a hemocytometer. Three-day-old seedlings with a radical of 1 to 1.5 cm long were immersed in the sporangial suspension and kept at 16°C overnight (Cohen and Sackston 1973). Inoculated seedlings were planted into trays containing clear moistened perlite (diameter = 4 mm) and grown in a growth chamber with a photoperiod of 12 h. The experiment was carried out twice with each isolate using 15 seeds per differential line with two replicates. Bidistilled water was sprayed on the plants 9 days after inoculation, and then trays were covered with a black polyethylene bag and maintained at 19°C overnight to induce sporulation. The first disease assessment was done based on cotyledons bearing white sporulation. Next, a second evaluation was performed 21 days after inoculation assessing stunting of the plants, chlorotic lesions on true leaves, and damping-off. All four isolates examined caused disease on differential lines HA-304, RHA-265, RHA-274, PMI-3, PM-17, and HA-335, whereas the other lines showed no symptoms and signs of sunflower downy mildew. As a result, it was concluded that the presence of *P. halstedii* pathotype 734 was confirmed. This pathotype is likely widespread in Hungary, because it could be detected from three different regions. Moreover, the possibility that pathotype 734 is present in Hungary has been raised before (Iwebor et al. 2018). This pathotype is already widespread in the United States and Russia and is considered to be highly aggressive, because it was able to infect hybrids with resistance genes *PI₆* and *PI₇* (Iwebor et al. 2018; Spring 2019). To our knowledge, this is the first report of pathotype 734 of *P. halstedii* in Hungary and Central Europe. Continuous monitoring and incorporation of new resistance genes into sunflower hybrids are essential steps in the future to control *P. halstedii*.

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