DOI: 10.1515/plass-2017-0021

Emil Stefańczyk

Plant Breeding and Acclimatization Institute – National Research Institute, Młochów
Research Center, Platanowa Str. 19, 05-831 Młochów, Poland;
e.stefanczyk@ihar.edu.pl

ASSESSMENT OF POTATO TUBER RESISTANCE AGAINST DRY ROT

ABSTRACT

Fungi of the genus *Fusarium* display inter- and intraspecific variation in their pathogenic capabilities. Although no resistance genes against dry rot have been identified, some indications of the possible defense responses are noticeable, since a single isolate might cause lesions of various sizes when infecting tubers of different potato cultivars. Pathogenicity tests are routinely used to assess potato tubers resistance against dry rot; a setup of pathogenicity test performed at Młochów Research Center is described in this work.

Key words: *Fusarium*, pathogenicity, *Solanum tuberosum*

INTRODUCTION

So far, no resistance genes against dry rot have been identified, however particular potato cultivars differ in their susceptibility which depends on the aggressiveness of the *Fusarium* species used. As shown in the study by Stefańczyk *et al.* (2016) *F. sambucinum* was the most pathogenic among the 12 different species used for inoculation of four potato cultivars. Interestingly, differences in pathogenic capabilities were also observed between isolates belonging to the same species. To assess potato tuber resistance against dry rot, highly aggressive *Fusarium* isolates, previously evaluated on selected cultivars, should be used (Sobkowiak and Śliwka, 2013). A high inoculum concentration leads to the appearance of earlier dry rot symptoms, thus allowing the assessment to be conducted after three weeks of incubation (Sobkowiak and Śliwka, 2013); this can help avoid secondary infections often caused by soft rot bacteria.

Pathogenicity tests are often described in literature, but no unified dry rot resistance assay has been presented. The differences cover inoculum preparation (conidial suspension, agar plugs with *Fusarium* mycelium), temperature (4°C, 10°C, 16°C, 20°C) and time (20, 21, 30, 35 days) of incubation or lesion size scoring (depth of necrosis, depth and width averages, lesion in mm², rot volume in mm³, 1-9 scale according to the percentage of the diseased area); common conditions comprise the absence of light and maintenance of high relative humidity during the incubation period (Gachango *et al.*, 2012; Gashgari and Gherbawy, 2013; Peters *et al.*, 2008a; Peters *et al.*, 2008b; Du *et al.*, 2012; Zarzycka, 2001). Instead of wounded tubers, tuber slices inoculated with inoculum plugs have also been proposed for pathogenicity tests (Desjardins and Gardner, 1989).

MATERIALS

- 1) Single-spore *Fusarium* spp. cultures isolated as described by Stefańczyk and Sobkowiak (2017)
- 2) Potato tubers
 - a) for assessing the resistance to dry rot
 - b) for inoculum and test conditions control
- 3) Metal rod
- 4) Pipette tips
- 5) Plastic trays with glass covers
- 6) Ruler

EQUIPMENT

- 1) Microscope (Carl Zeiss Microscopy, Jena)
- 2) Haemocytometer (Marienfeld Superior, Thoma)
- 3) Pipettes
- 4) Room with controlled temperature

PROCEDURE

The inoculum preparation

- 1) Single-spore fungal cultures grown on a PSA medium at 16°C for three weeks are washed with sterile tap water.
- 2) Spore concentration is adjusted to $2.5 \times 10^6 \times \text{ml}^{-1}$ using a haemocytometer.

Pathogenicity test

- 1) The potato tubers are wounded on their apical ends with a tool (a metal rod) causing a wound 10 mm in depth and 5 mm in diameter (Photo 1).

Three to ten tubers of each assessed potato genotype are usually tested per each of the two replicates.

Photo 1. The tool used for wounding the potato tubers in the pathogenicity test. The wound is then inoculated with *Fusarium* sp. inoculum introduced with a pipette.

Note: It is advised to test at least three tubers in each replication, due to the variability of the infection process. At Młochów Research Center, five tubers per replication are tested.

- 2) The wounds are inoculated with 50 μ l of inoculum using a pipette and the tubers are incubated for 3 weeks in a plastic tray in the dark at 16°C. To maintain the humidity, glass is used as a cover. As a control of the inoculum and of the test conditions, tubers of standard cultivars of known susceptibility are tested simultaneously (Photo 2).

Photo 2. Tubers of cv. Bartek inoculated with (from left to right) *F. avenaceum*, *F. graminearum*, *F. sambucinum* and *F. solani* isolates after three weeks of incubation in the pathogenicity test.

Note: The incubation period may have to be adjusted according to size of the tubers and the aggressiveness of the isolate.

At Młochów Research Center, four standard cultivars are used, among which cvs. Gawin and Hinga exhibit phenotypes resistant and susceptible to dry rot, respectively.

- 3) The tubers are cut in half along their longer axes after the incubation period. Using a ruler, two perpendicular diameters of each lesion are measured and their mean value denotes the lesion size.

Note: For your own safety, consider use of a procedure mask during this step.

ACKNOWLEDGEMENTS

This work was financed by a statutory project No 1-3-00-3-06 from the Plant Breeding and Acclimatization Institute, Poland.

REFERENCES

- Desjardins A.E., Gardner H.W. 1989. Genetic analysis in *Gibberella pulicaris*: rishitin tolerance, rishitin metabolism, and virulence on potato tubers. *Mol. Plant Mic. Inter.* 2: 26-34.
- Du M., Ren X., Sun Q., Wang Y., Zhang R. 2012. Characterization of *Fusarium* spp. causing potato dry rot in China and susceptibility evaluation of Chinese potato germplasm to the pathogen. *Potato Res.* 55: 175–184.
- Gachango E., Hanson L.E., Rojas A., Hao J.J., Kirk W.W. 2012. *Fusarium* spp. causing dry rot of seed potato tubers in Michigan and their sensitivity to fungicides. *Plant Dis.* 96: 1767–1774.
- Gashgari R.M., Gherbawy Y.A. 2013. Pathogenicity of some *Fusarium* species associated with superficial blemishes of potato tubers. *Pol. J. Microbiol.* 62: 59-66.
- Peters R.D., MacLeod C., Seifert K.A., Martin R.A., Hale L.R., Grau C.R., MacInnis S. 2008a. Pathogenicity to potato tubers of *Fusarium* spp. isolated from potato, cereal and forage crops. *Am. J. Potato Res.* 85: 367–374.
- Peters J.C., Lees A.K., Cullen D.W., Sullivan L., Stroud G.P., Cunnington A.C. 2008b. Characterisation of *Fusarium* spp. responsible for causing dry rot of potato in Great Britain. *Plant Pathol.* 57: 262–271.
- Sobkowiak S., Śliwka J. 2013. Grzyby z rodzaju *Fusarium* powodujące suchą zgniliznę bulw ziemniaka. *Ziemniak Polski* 4: 18-24.
- Stefańczyk E., Sobkowiak S., Brylińska M., Śliwka J. 2016. Diversity of *Fusarium* spp. associated with dry rot potato tubers in Poland. *Eur. J. Plant Pathol.* 145: 871-884.
- Stefańczyk E., Sobkowiak S. 2017. Isolation, identification and preservation of *Fusarium* spp. causing dry rot of potato tubers. *Plant Breed. Seed Sci.* 76: 45-51
- Zarzycka H. 2001. Assessment of potato tuber resistance against soft rot and dry rot. In: E. Zimnoch-Guzowska, J. Syller, M. Sieczka (Eds.), *The methods of evaluation and selection applied in potato research and breeding*. Plant Breeding and Acclimatization Institute, Radzików. Monografie i Rozprawy Naukowe IHAR Radzików. 10a: 88–9.