Disease Note

FIRST REPORT OF *IRIS MILD MOSAIC VIRUS* FROM *IRIS XIPHIUM* IN IRAN

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Iris mild mosaic virus (IMMV, genus *Potyvirus*, family *Potyviridae)* has been previously reported in *Iris* species worldwide (Brunt, 1986). Since November 2015, a total of 85 symptomatic leaf samples of bulbous *Iris xiphium* plants showing mild mosaic and chlorosis were collected from greenhouses of four different provinces of Iran. 94% of collected samples reacted positively with genus-specific monoclonal antibodies for potyviruses (AS-0573/1 DSMZ, Germany) in antigen coated-plate ELISA (Jordan and Hammond, 1991). Crude leaf extracts of 15 symptomatic potyvirus infected *Iris* plants were individually mechanically inoculated on nine seedlings (three replications of three plants each) of 16 different indicator plant species belonging to Chenopodiaceae, Cucurbitaceae, Fabaceae and Solanaceae families. No symptoms developed on inoculated test plants except for *Chenopodium quinoa* in which numerous chlorotic local lesions were induced by all inoculated isolates. Total RNA was extracted from several local lesions of five *C. quinoa* leaves and used in RT-PCR assay with a new specific primer pair, IMMV-F/IMMV-R (5'-CGAGATACGGAAGTTCTACGCA-3'/5'-GGGTTGTGCGTAATCAAGTAGTGG-3'), which was designed based on nucleotide sequence of IMMV-WA-1 isolate (GenBank accession No. JF320812) to amplify a *ca.* 700 bp region encompassing the partial nuclear inclusion protein b and coat protein (3'-NIb, 211 bp /5'-CP, 492 bp) genes of IMMV genome. Following cloning and sequencing, BLAST analysis of an isolate (IMMV-Ar, KX870019) sequence showed the highest range of identity (82-99%) with existing IMMV sequences, including isolates DC4b (99%, DQ436919; New Zealand) and Bate2 (98%, JN127338; Australia). Phylogenetic analysis based on partial NIb-CP sequences of all five available IMMV isolates showed that IMMV-Ar clustered with isolates Bate2 and DC4b. To our knowledge, this is the first report of IMMV occurrence on *Iris xiphium* from Iran.

Brunt A.A., 1986. *Iris mild mosaic virus*. CMI/AAB Descriptions of Plant Viruses, No. 324 http://www.dpvweb.net.

Jordan R., Hammond J., 1991. Comparison and differentiation of potyvirus isolates and identification of strain-, virus-, subgroup-specific and potyvirus group-common epitopes using monoclonal antibodies. *Journal of General Virology* **72**: 25-36.

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Disease Note

FIRST REPORT OF *DIPLODIA SERIATA* AS CAUSAL AGENT OF ALMOND TREE BRANCH DIEBACK IN TUNISIA

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A branch dieback of almond trees (cv. Achaak) was noticed in 2016 in the region of Sfax. 10% of trees showed dead branches with sunken areas. Infected tissues were abnormally dark and cross sections revealed a light-brown and irregularly shaped canker developing from the graft-union. Numerous pycnidia were observed on the surface of infected branches with severe gummosis on the buds. To identify the pathogen, 35 small pieces of infected tissues were plated on potato dextrose agar. After incubation at 25°C for 5 days, 15 isolates were consistently isolated. Colonies were grayish white, producing dark melanised hyphae and pycnidia after 10 days of incubation. Conidia were first hyaline, and then turned brown as they matured. They were aseptate, rounded at both ends with thick melanised cell walls, 19 to 25.7×9.5 to 11.03 µm. Based on colony characteristics, all isolates were identified as *Diplodia seriata* (Phillips *et al*., 2007). Molecular identification was performed by sequencing the ITS, the EF-1-α and the β-tubulin genes. BLAST searches of ITS (KY275259), EF-1-α (KY284865) and β-tubulin (KY275260) sequences, respectively revealed 98%, 99% and 100% identity to *Diplodia seriata*. Pathogenicity tests were performed on detached stems, wounded with a scalpel and inoculated with mycelial plugs (Yan *et al*., 2011). Controls were inoculated with sterile agar plugs. Inoculated stems were placed in polyethylene boxes and incubated at 25°C for 30 days. While mock-infected stems remained healthy, inoculated stems developed brown discoloration, with small dark pycnidia growing on the surface. The pathogen was isolated from inoculated stems, thus fulfilling Koch's postulates. To our knowledge, this is the first report of canker disease caused by *Diplodia seriata* in Tunisia.

Phillips A.J.L., Crous P.W, Alves A., 2007. *Diplodia seriata*, the anamorph of "*Botryosphaeria obtusa*". *Fungal Diversity* **25**: 141-155

Yan J.Y., Peng Y.L, Xie Y., 2011. First Report of Grapevine Trunk Disease Caused by *Botryosphaeria obtusa* in China. *Plant disease* **95**: 616

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