

DISEASE NOTE

FIRST REPORT OF *ASPERGILLUS WELWITSCHIAE* AS A POSTHARVEST PATHOGEN OF *BRASSICA CAMPESTRIS* SEEDS IN PAKISTAN

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Mustard (*Brassica campestris*), one of the vegetables grown in Pakistan, is a highly valued crop also because of its edible oil-rich seeds, that have great medicinal potential. Mustard seeds stored in Lahore were found to be colonized by a previously undetected black powdery mold. Twenty infected seeds from three different warehouses of Lahore were collected and spores present on the seeds were transferred aseptically to Czapek dox agar (CZA) and incubated at 25°C. A 7-day-old pure culture was used for the identification of the recovered fungus. Colonies were black, 4 cm in diameter and showed abundant radiatel heads bearing brown conidia 4-5.5 µm in diameter. Metulae were 13 × 3.5 µm and phialides 10 × 3.5 µm in size. Identification of pathogen was carried out by sequencing the ITS region of rDNA and part of the calmodulin gene (CAL) (White *et al.*, 2000). BLAST results showed 99% similarity of the ITS sequence (GenBank accession No. KT277269) and 100% similarity of the calmodulin gene (LN890507) to strain DTO_178C2 and PW3050 of *Aspergillus welwitschiae*. To determine the pathogenicity of the fungus, 50 randomly selected seeds were surface-sterilized, soaked in a spore suspension (10⁴ spores/ml) for 30 sec and, after drying, were transferred aseptically onto a layer of sterilized moist blotting papers. Control seeds were treated with sterilized water. All Petri dishes were incubated at 25°C. After 7 days of incubation, 75% of the seeds were colonized by the pathogen and failed to germinate while all control seeds germinated to produce healthy seedlings. To the best of our knowledge, this is the first report of *A. welwitschiae* from Pakistan causing postharvest rot of mustard seeds.

White T.J., Bruns T., Lee S., 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Taylor J., Innis A., Gelfand D.H., Sninsky J.J. (eds). PCR Protocols, pp. 315-322. Academic Press, San Diego, CA, USA.

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FIRST REPORT OF *ALTERNARIA CUCURBITAE* CAUSING BUNCH ROT ON GRAPE IN IRAN

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In July 2014, symptoms of berry cracking and black rot were observed on bunches of vines grown in a vineyard in the province of Hamedan (Iran). A fungus was successfully isolated from symptomatic berries plated on potato dextrose agar (PDA) incubated at 23°C. Colonies were brownish green with concentric rings, hyphae had pale yellow to brown septa and conidiophores were golden-brown, short, geniculate and sympodial. Conidia (18-20 × 8-10 µm) were mostly broadly obovoid to subspherical with longitudinal septa, mature conidia were primarily subspherical with few diagnostic obovoid conidia (Woudenberg *et al.*, 2013). Based on these morphological traits, the pathogen was identified as *Alternaria cucurbitae*. Molecular identification was based on amplification of the β-tubulin gene sequence (Woudenberg *et al.*, 2013). The 550 bp product (GenBank accession No. KU324181), shared 99% similarity with the comparable sequence of *A. cucurbitae* [= *Ulocladium cucurbitae* (JQ67200101)]. A pathogenicity test was conducted on detached grapevine bunches (13-15 cm in length). Each sterile Petri dish contained 3 small clusters at the postveraison stage, each with 10 berries, which were sprayed with a spore suspension (1 × 10⁶ spores/ml), while control samples were sprayed with sterile water. Bunch rot symptoms appeared after 7 days incubation at 25°C. *A. cucurbitae* was successfully re-isolated from artificially infected berries while the controls remained symptomless. *A. cucurbitae* has been previously reported as the agent of leaf spot on cucumber in California (Vakalounakis, 1990). To our knowledge, this is the first report of *A. cucurbitae* as a bunch rot agent on grape in Iran and possibly worldwide.

Vakalounakis D.J., 1990. Host range of *Alternaria alternata* f. sp. *cucurbitae* causing leaf spot of cucumber. *Plant Disease* **74**: 227-230

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