Disease Note

FIRST REPORT OF *ONION YELLOW DWARF VIRUS* AND *GARLIC COMMON LATENT VIRUS* INFECTION IN GARLIC FROM NEPAL

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Garlic (*Allium sativum* L.) is one of the most important culinary herbs in the Indian subcontinent. Several viruses belonging to the genera *Potyvirus*, *Carlavirus* and *Allexivirus* are known to infect garlic worldwide (Dijk, 1994; Walkey and Antill, 1989). Leaves from 20 different samples of cultivar 'Sauntha lasoon' showing mild to severe mosaic symptoms were collected in April of 2013 from two fields of Dharchula, Nepal. Direct antigen coated (DAC)-ELISA was performed with antisera to *Onion yellow dwarf virus* (OYDV), *Shallot latent virus* (SLV) and *Garlic common latent virus* (GarCLV) (Bioreba, Reinach, Switzerland). All the samples were positive for OYDV and 16 were positive for GarCLV. These results were confirmed by reverse transcription (RT)-PCR using specific primers (Majumder and Baranwal, 2014) and total RNA extracted from 100 mg of leaves with the RNeasy Plant Mini kit (Qiagen, USA) according to the manufacturer's protocol. Expected amplicons of *ca.* 320 bp for OYDV and *ca.* 450 bp for GarCLV were obtained from all the samples tested, indicating mixed infections. Direct sequencing of the PCR products produced 276 bp and 461 bp long nucleotide sequences with 78% and 91% identity with sequences of an OYDV isolate from garlic in India (GenBank accession No. DQ519034) and a GarCLV isolate from garlic in India (GenBank accession No. FJ154841), respectively. SLV was not found by ELISA or RT-PCR in any of the samples tested. To our knowledge, this is the first report of OYDV and GarCLV in garlic in Nepal.

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Disease Note

FIRST REPORT OF DIEBACK OF OLIVE TREES CAUSED BY *PHOMA FUNGICOLA* IN TUNISIA

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During spring 2013, a new disease was observed on olive (*Olea europea*) cv. Chemlali in orchard in Sahlin (Sahel of Tunisia). Symptoms included abundant dead branches and wilted leaves and shoot necrosis. Shoots showing dieback symptoms were disinfected with 2% sodium hypochlorite, rinsed in sterile distilled water and air dried. Several fragments (3 x 3 mm) of infected shoots were cut and placed on potato dextrose agar medium (PDA). All plates were incubated at 25°C for 4 days under continuous fluorescent light. A pycnidial fungus was consistently isolated from branch cankers and identified as *Phoma fungicola* Aveskamp, Gruyter et Verkley, based on morphological characteristics and analysis of the ITS gene region (White *et al*., 1990). The sequence showed high identity (99%) with a reference sequence (strain H11 H10; accession No. KF29376 3.1). Pathogenicity tests were conducted on 2-year old olive plants (cv. Chemlali). A mycelial plug cut from the margin of an actively growing colony of the fungus was placed into a shallow wound (0.4 cm^2) inferted with a sterilized scalpel on the stem base. Inoculated wounds were wrapped with Parafilm. In control plants, sterile PDA plugs were placed into artificial wounds. Ten replicate inoculated plants were used and maintained in a greenhouse at 25°C. Two months after the inoculation, the inoculated trees reproduced stem browning symptoms observed in the field, while control plants remained healthy. Koch's postulates were then verified and *P. fungicola* was isolated from inoculated stems, whereas the controls were free of the fungus.

Phoma sp. and *P. incompta* have been reported as responsible for branch dieback of olive tree in Tunisia and Italy, respectively (Rhouma *et al*., 2010; Ivic *et al*., 2010). To the best of our knowledge, this is the first report of *P. fungicola* as a causal agent of dieback of olive trees in Tunisia.

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