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SUSTAINABLE AGRICULTURE AND RURAL DEVELOPMENT IV

Book of Abstracts



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HEALTHY SOIL - HEALTHY PLANT: CONTAMINATED SOIL AS A SOURCE OF INOCULUM OF *FUSARIUM VERTICILLIOIDES* (SACC.) NIRENBERG

Maja Ignjatov¹, Dragana Milošević¹, Janko Červenski¹, Slobodan Vlajić¹,
Boris Adamović², Snežana Jakšić¹, Đorđe Vojnović²

Abstract

A wide occurrence of wilt and rot caused by *Fusarium* species has been observed in the last few years in many vegetable-growing areas in the Republic of Serbia. Symptoms of rot and wilt of celery, garlic, and onion were noticed and after determination, *Fusarium* spp. was identified as the main causal agent of the disease. Most of the *Fusarium* species that cause disease on vegetable crops (garlic, onion, celery) overwinter in soil debris and soil. This study was conducted to isolate and identify *Fusarium* from soil samples by conventional and molecular methods. Soil samples were taken under the diseased plant, and 25 samples were collected in order to perform isolation of *Fusarium* species that cause rot and wilt during production. Each soil sample (10 g) was transferred to a glass bottle with 100 ml of sterile distilled water and put in a shaker for 30 min. An extract (0.2 ml) from each sample was placed on the bottom of the Petri dish and then a liquid medium of potato dextrose agar – PDA, was added and incubated at 28°C. After seven days all isolates were examined morphologically and re-isolated completing Koch's postulates. Colony morphology was recorded from cultures grown on PDA and CLA. Nine isolates, when grown on PDA, develop fast-growing, abundant, vinaceous aerial mycelium-producing dark violet pigments. Only microconidia are attached to each other in long chains. Macroconidia and chlamydospores were absent. Based on the morphological characteristics, compared with positive control and literature description isolates were identified as *Fusarium verticillioides* (Sacc.) Nirenberg. To perform molecular confirmation, total genomic DNA was extracted from the mycelium with a DNeasy Plant Mini Kit (Qiagen, Hilden, Germany). Following DNA extraction, the translation elongation factor *TEF1*-alpha region was amplified by PCR with the primer pair *EF1* and *EF2*. A positive control isolates previously

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identified as *F. verticillioides* (NCBI code MH496027) was used. The sequences were compared with those in GenBank. The *TEF1*-alpha gene sequences of the Serbian isolates showed the highest identity of 100% with a Argentinian isolate (OR529768) and South African *F. verticillioides* isolates Accession No.: OR498563, OR498540 and OR498543-44. These species can infect vegetable plants through, contaminated seeds, or through spores carried over by wind, water, and insects. *F. verticillioides* is a well-known soil- and seed-borne facultative endophyte and can enter systemically through the root system from the soil. Hence, this species is known to produce mycotoxins severe for human health and monitoring in Serbia will be continued.

Key words: *Fusarium verticillioides*, soil, *TEF1*-alpha gene, sequencing.

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