Title First report of in vitro fludioxonil-resistant isolates of *Fusarium* spp. causing potato dry rot in Michigan

Authors E. Gachango, W. Kirk, L. Hanson, A. Rojas, P. Tumbalam and K. Shetty

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Abstract

Fusarium dry rot of potato (*Solanum tuberosum*) is a postharvest disease caused by several *Fusarium* spp. Dry rot is managed primarily by reducing tuber bruising and promoting rapid wound healing. Dry rot symptomatic tubers were collected from Michigan seed lots in 2009 and 2010. The isolates may not have been exposed to fludioxonil because currently applications are restricted to seed not intended for seed production (3). Small sections were cut from the margins of necrotic regions with a scalpel, surface sterile in 10% sodium hypochlorite for 10 s, rinsed twice in sterile distilled water, and blotted with sterile filter paper. The tissue pieces were plated on half-strength potato dextrose agar (PDA) amended with 0.5 g/liter of streptomycin sulfate. The dishes were incubated at 23°C for 5 to 7 days. Cultures resembling *Fusarium* spp. were transferred onto water agar and hyphal tips from the margin of actively growing isolates were removed with a sterile probe and plated either on carnation leaf agar (CLA) or on half-strength PDA to generate pure cultures. Fusarium isolates were obtained and used for further studies. Among them, 54 were identified as *Fusarium oxysporum* and 23 as *F. sambucinum.* Identification was based on colony and conidial morphology on PDA and CLA, respectively. The identity was confirmed through DNA extraction followed by amplification and sequencing of the translation elongation factor (EF-1α) gene region. The Fusarium-ID v. (2) and the NCBI database were used to obtain the closest match to previously sequenced materials. Pathogenicity testing was done on disease-free potato tubers, cv. FL 1879. Tubers were surface sterilized for 10 min in 10% sodium hypochlorite and rinsed twice in distilled water. Three tubers per isolate were injected with 20 µl of a conidial suspension (10^6 conidia/ml) made from cultures grown on PDA for 7 days. Control tubers were injected with 20 µl of sterile distilled water. All tubers inoculated with *F. sambucinum* and *F. oxysporum* developed typical potato dry rot symptoms consisting of dry brown decay lesions. *F. sambucinum* and *F. oxysporum* were reisolated from all symptomatic tubers. An effective concentration for 50% reduction in growth (EC_{50}) was determined for each *F. sambucinum* and *F. oxysporum* isolate for thiabendazole (TBZ), fludioxonil, and

difenoconazole using the spiral gradient endpoint method (1). Sensitive and resistant *F. sambucinum* and *F. oxysporum* isolates were reported. Fifteen isolates of *F. sambucinum* and thirty-four of *F. oxysporum* were resistant to fludioxonil with EC_{50} greater than 130 mg/liter. The remainder was sensitive to fludioxonil with EC_{50} ranging from 0.8 to 4.9 mg/liter. To our knowledge, this is the first report of resistance to fludioxonil in isolates of *F. sambucinum* and *F. oxysporum* in Michigan. Fusarium insensitivity in laboratory studies may not translate directly to commercial production. This disparity may result from interactions not experienced in mixed populations or within a living host. There has been no compelling evidence to suggest that fludioxonil has failed to perform because of insensitivity to Fusarium. The occurrence of such isolated strains necessitates the development and registration of partner chemistries that can preempt any future concerns on lack of performance of products in use.