

DIFFERENCES IN GENE EXPRESSION BETWEEN PATHOGENIC AND BIOCONTROL FUSARIUM

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Research is being conducted to identify genes involved in biocontrol of Fusarium wilt by nonpathogenic Fusaria, and to understand the genetic relationship between biocontrol and other Fusaria. In previous work, 415 *Fusarium oxysporum* were isolated from one conventional and one organic farm in Florida. These were screened in the greenhouse for pathogenicity on Bonny Best tomato. Of these, 26 caused wilt (*F. oxysporum* f. sp. *lycopersici*) and 53 caused root and crown rot (*F. oxysporum* f. sp. *radicis-lycopersici*). The remaining 336 isolates were considered nonpathogenic on tomato. To determine race of the pathogen, the 26 *F. o. lycopersici* were tested on different tomato cultivars and 15 isolates were identified as Race 1, 6 as Race 2 and 5 as Race 3.

The 336 nonpathogenic strains were screened in the greenhouse for their ability to control Fusarium wilt on tomato. Tomatoes were drenched at seeding and one day prior to transplant (4 weeks after seeding) with 5 ml of 10^5 spores (macroconidia + microconidia + chlamydospores) / ml. Plants were transplanted into field soil infested with 10^3 spores/g soil (at least 20% chlamydospores) of *F. o. lycopersici* Race 1. Five plants in a single pot were considered one replicate and treatments were replicated four times. The experiment was repeated. After five weeks, disease was assessed three ways: disease severity rating (0 to 5 with 0 = no symptoms and 5 = dead plant), incidence (stem plating on a selective medium), and fresh weight of the shoots. Isolates were not clearly divided into biocontrol or non-biocontrol strains. Rather, there was continuum of abilities. Strains were grouped into high, medium or low biocontrol ability.

Differences in gene expression were studied using differential display analysis. RNA was extracted from two isolates identified as pathogens and two isolates with high biocontrol ability. First strand cDNA was amplified from total RNA by reverse transcription. The cDNA was amplified again using anchored and random primers and the resulting products were separated by gel electrophoresis. The fragments were visualized using a fluorescent scanner. Twenty-two cDNA bands with potential differential expression between the pathogenic and biocontrol strains were identified. These bands were excised and the cDNAs sequenced. Bands showing high homology to known sequences included an ATP synthase, an ABC transporter, and a lysine permease. ABC transporters can function in detoxification of toxic compounds and have not been previously reported in Fusarium. Studies are currently underway with additional pathogenic, biocontrol and saprophytic strains to determine whether these genes are present in the different phenotypes (but expressed differently), as well as studies on the regulation of the ABC transporter.

