Available online at www.sciencedirect.com
SCIENCE DIRECT®
Biological Control 34 (2005) 165–169
Biological Control
www.elsevier.com/locate/ybcon

Compatibility of the biocontrol fungus *Fusarium oxysporum* strain CS-20 with selected fungicides

D.R. Fravel *, K.L. Deahl, J.R. Stommel

Vegetable Laboratory, United States Department of Agriculture, Agricultural Research Service, The Henry A. Wallace Beltsville Agricultural Research Center—West, Beltsville, MA 20705, USA

Received 31 December 2004; accepted 18 April 2005
Available online 26 May 2005

Abstract

Fusarium oxysporum strain CS-20 reduces incidence of Fusarium wilt in tomato and other crops. To be integrated into most production systems, strain CS-20 must be compatible with other management practices. We assessed the compatibility of strain CS-20 with seven fungicides recommended for tomato in Maryland. Radial growth of strain CS-20 was recorded on agar medium amended with various concentrations of the fungicides. Fungicides tested did not kill strain CS-20 at the concentrations tested in the in vitro experiment. Azoxystrobin (Quadris) and chlorothalonil (Bravo) were most toxic to strain CS-20 and significantly reduced growth rate and final colony size at 10 ppm a.i. or greater concentrations compared to growth on unamended medium. Thiram (thiram) significantly reduced final colony size at 30 ppm or greater. Mefenoxam + chlorothalonil (Ridomil Gold Bravo) significantly reduced final colony size at 50 ppm or greater. Mancozeb (Manzate) and mancozeb + copper (Mankocide) reduced final colony size only at 100 ppm, while mefenoxam (Ridomil Gold) and mefenoxam + copper (Ridomil Gold Copper) did not affect growth of strain CS-20. In greenhouse tests, tomatoes were drenched with strain CS-20 at seeding and just before transplanting into field soil infested with the pathogen. Plants were treated with fungicides at the highest label rate. Mancozeb is labeled as both a seed treatment and a spray, and was applied each of these ways as separate treatments. Disease incidence of plants from seeds treated with thiram and strain CS-20 was not different from those in the pathogen only control, indicating that thiram was toxic to strain CS-20. Other fungicides toxic in vitro, were less toxic in greenhouse tests, probably because they are applied as sprays to the above-ground portions of the plant. Published by Elsevier Inc.

Keywords: *Fusarium oxysporum*; Fusarium wilt; Biopesticide; Azoxystrobin; Chlorothalonil; Mancozeb; Mefenoxam; Thiram

1. Introduction

Development of alternative control strategies such as biocontrol can provide additional management tools as supplements to other control measures for plant diseases, for use in rotation with other control measures, or as back-up when favored control measures are withdrawn from the market or fail due to new strains or races of the pathogen.

Fusarium wilt, caused by *F. oxysporum* f. sp. *lycopersici*, is a serious problem for tomato production in many areas. The disease is managed through resistant cultivars and pre-plant soil fumigation. Because new races of the pathogen can develop and fumigants are being lost from the market, alternative control strategies are being investigated. There are many examples in the literature of using nonpathogenic *Fusarium* spp. to control Fusarium wilts (reviewed in Fravel et al., 2003). In field and greenhouse studies, *F. oxysporum* strain CS-20 has demonstrated potential for reducing the incidence of Fusarium wilt on tomato, muskmelon and basil (Larkin and Fravel, 1998, 1999a,b, 2002a,b; Larkin et al., 1999).

* Corresponding author. Fax: +1 301 504 5555.
E-mail address: fraveld@ba.ars.usda.gov (D.R. Fravel).

1049-9644/$ - see front matter. Published by Elsevier Inc.
doi:10.1016/j.biocontrol.2005.04.007

To successfully integrate biocontrol organisms into production systems, the compatibility of these organisms with pesticides used in the production system must be known. This work was undertaken to determine the compatibility of *F. oxysporum* strain CS-20 with fungicides recommended for use on tomato in Maryland.

2. Materials and methods

2.1. In vitro experiments

The biocontrol strain *F. oxysporum* strain CS-20 was originally isolated in Florida from a soil suppressive to Fusarium wilt of watermelon (Larkin et al., 1996). For in vitro experiments, the fungus was grown on potato dextrose agar (PDA; Sigma, St. Louis, MO) at 25 °C for 1 week. One-half centimeter disks from the growing edge of the fungal colony were transferred to the center of fresh PDA amended to result in 0, 10, 30, 50, or 100 ppm a.i. of each of the following fungicides: azoxystrobin (Quadris; Syngenta, Greensboro, NC); chlorothalonil (Bravo Ultrex; Syngenta); mancozeb (Manzate 75DF; Griffin, Valdosta, GA); mancozeb + copper (Mankocide DF; Griffin); mefenoxam (Ridomil Gold EC; Syngenta); mefenoxam + chlorothalonil (Ridomil Gold Bravo, Syngenta); mefenoxam + copper (Ridomil Gold Copper, Syngenta); and thiram (Thiram 50WP Dyed; Gustafson, Plano, TX) (Table 1). Active ingredient (a.i.) refers to the sum of the two active ingredients when two are present in the fungicide as it is sold. Agar was amended by preparing 100 ml of PDA and adding 1 ml of sterile distilled water (SDW) or the appropriate fungicide dilution brought up to 1 ml with SDW in sterile Eppendorf tubes. Eighteen milliliters of medium was dispensed into each of four replicate 9-cm diameter plastic petri plates. Two perpendicular lines were drawn on the bottom of each petri plate and the fungal disk was placed at the intersection of the two lines.

The diameter of the fungus was measured on each of the two perpendicular lines drawn on the plate every 2–3 days until the fungus reached the edge of the plate (up to 14 days). Data were analyzed by a general linear model (proc glm, SAS, Cary, NC). Growth rate was calculated. The experiment was repeated once.

2.2. Greenhouse experiments

Inoculum of strain CS-20 and the pathogen, *F. oxysporum* f. sp. *lycopersici* race 1, were grown in an aqueous suspension of 1%(w:v) soy hull at 25 °C with shaking (100 rpm) for 2–4 weeks before use (Hebbar et al., 1996). Spores (microconidia + macroconidia + chlamydospores) were enumerated with a hemacytometer and spore concentrations were adjusted with SDW.

For fungicides labeled for seed use (manzate, thiram), tomato seeds (*Lycopersicon esculentum* cv. Bonny Best) were treated as described on the product label. Treated or nontreated seed were planted in soilless potting mix (Redi-Earth; Scotts-Sierra, Marysville, OH). Five seeds were planted in each cell of plastic seedling trays (98 cells, $3.3 \times 3.3 \times 5$ cm tall). One cell with five seeds was considered a replicate. Each replicate was drenched with 5 ml of a 10^6 spores/ml aqueous suspension of strain CS-20. After 6 weeks, each replicate was again drenched with 5 ml of a 10^6 spores/ml aqueous suspension of strain CS-20. The following day, each replicate (4–5 plants) was transplanted into a 15 cm diameter plastic pot with nonsterile field soil (Galeston gravely sandy loam) infested with 10^4 spores/g soil of the pathogen. Treatments were replicated 10 times. Plants were sprayed at 1, 3, and 5 weeks after transplant with the maximum label rate of azoxystrobin, chlorothalonil,

Table 1
Label rates, chemical components, chemical class, and mode of action of fungicides used in experiments

Fungicide	Highest label rate	Component	Chemical class	Mode of action
Bravo Ultrex	2.40 kg a.i./ha	Chlorothalonil	Substituted benzene chloronitrile	Combines with NH_2 or SH group of essential metabolic compounds
Manzate 75DF	2.52 kg a.i./ha	Mancozeb	Ethylene bisdithiocarbamate	Inactivates SH groups in amino acids
Mankocide DF	3.42 kg a.i./ha	Mancozeb	Ethylene bisdithiocarbamate	Inactivates SH groups in amino acids
		Copper hydroxide	Copper	Nonspecific denaturization of protein
Ridomil Gold Bravo	1.46 kg a.i./ha	Mefenoxam	Phenalymide	Depresses nucleic acid synthesis
		Chlorothalonil	Substituted benzene	Combines with NH_2 or
			Substituted aromatic	SH group of essential metabolic compounds
Ridomil Gold Copper	1.46 kg a.i./ha	Mefenoxam	Phenalymide	Depresses nucleic acid synthesis
		Copper	Copper	Nonspecific denaturization of protein
Ridomil Gold EC	1.17 L a.i./ha	Mefenoxam	Phenalymide	Depresses nucleic acid synthesis
Thiram 50WP Dyed	1.87 g a.i./kg seed	Thiram	Dithiocarbamate	Interferes with oxygen uptake and inhibition of sulfur containing enzymes
Quadris	103.76 ml a.i./ha; 112.09 g a.i./ha	Azoxystrobin	Pyridimine	Inhibits biosynthesis; sterols