Comparative Studies with Two *Geotrichum* Species Inciting Postharvest Decays of Tomato Fruit

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ABSTRACT

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Growth rates of *Geotrichum candidum* and *G. penicillatum* (ATCC 48024) were compared on mature green and red tomato fruit at 5, 10, 15, and 20 C and on potato-dextrose agar (PDA) at 5, 10, 15, 20, 25, 30, 35, and 40 C. The rate of infection of mature green fruit by *G. penicillatum* was less than that by *G. candidum* at all temperatures. Both fungi grew at the same rate on red fruit. *G. penicillatum* was less aggressive and had a slower growth rate than *G. candidum* on PDA at temperatures from 10 to 30 C. Fungicidal effects of several chemicals were studied on mature green and red fruit inoculated with spore suspensions of the two species. Sodium hypochlorite, benomyl, thiophanate methyl, and vinclozolin retarded growth of *G. penicillatum* on green fruit; imazalil and ferbam retarded lesion development on red fruit. Sodium bicarbonate and potassium sorbate retarded *G. candidum* growth on green fruit, and thiophanate methyl and sodium bicarbonate retarded fungal growth on red fruit. None of these fungicides, however, prevented decay caused by either fungus. This is the first reported incidence of *G. penicillatum* causing postharvest decay of tomatoes.

Additional key words: sour rot, storage, watery rot

Sour rot of tomato (*Lycopersicon esculentum* Mill.) was first described by Pritchard and Porte in 1922 (17). *Geotrichum candidum* Link: Pers. has been shown to cause a sour rot of several vegetables (4,11) and fruits (3,4,10,13,16). This fungus is a wound pathogen, requiring injury for entry, and a common saprophyte easily recovered from stagnant water and wet soil (1,5,6,12, 13,16).

Symptoms of infection with *G. candidum* include brown water-soaked lesions that contain white mycelium. As decay progresses, fruit become soft and unmarketable. A distinctive sour odor is associated with the disease (14).

In 1978, an atypical watery rot of tomato was observed on fruit purchased at the Washington, DC, area wholesale market (15). It lacked the distinctive sour odor that accompanies decay caused by *G. candidum*. Mycelial growth on decayed tomatoes was somewhat different and arthrospores were slightly shorter and more rounded than those observed in *G. candidum* cultures. The pathogen of the atypical rot was isolated and subsequently identified as *G. penicillatum* (do Carmo-Sousa) Arx (5,10,17,18) by mycologists at the American Type Culture Collection, Rockville, MD. This is the first report of *G. penicillatum* causing postharvest decay of tomatoes.

This study compares growth rates of two representative isolates of *G. candidum* and *G. penicillatum* on potato-dextrose agar (PDA) and pathogenicity on mature green and red tomato fruit. *G. candidum* is a difficult pathogen to control because it is quite tolerant of many chemicals (7–9,11,16,19). Most fungicides have had little effect on decay in vitro. Those that have shown promise for the control of sour rot have made the fruit susceptible to other diseases (8). These results were the basis for our fungicide trials with *G. candidum* and *G. penicillatum*.

MATERIALS AND METHODS

One hundred ten red and 110 mature green tomatoes of uniform size (65–70 mm diameter) and color were selected for pathogenicity tests of *G. candidum* and *G. penicillatum*. Fruit were divided into two groups of 55 red and 55 mature green tomatoes each to serve as replicates. Twenty-five red and 25 green surface-sterilized tomatoes were inoculated with *G. candidum* in each of their four quadrants by puncturing with a spore-laden needle. Another 25 red and 25 green tomatoes were inoculated with *G. penicillatum* as indicated. Five red and five mature green tomatoes from each of the two replicates punctured with a sterile inoculating needle served as uninoculated controls. Treatments of five red or green inoculated tomatoes each were placed at 5, 10, 15, 20, and 25 C. Lesion diameter was measured daily.

Growth of *G. candidum* and *G. penicillatum* was compared on PDA. Media was added to 160 (15 × 100 mm) disposable petri plates, which were divided into two replicates of 80 each. Each plate was divided into four numbered quadrants. Forty plates in each replicate were inoculated with *G. candidum* and 40 inoculated with *G. penicillatum* along the edges of the plates so radial growth could be measured. Plates inoculated with fungal isolates were divided into eight treatments of five

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plates each. Treatments were placed at 5, 10, 15, 20, 25, 30, 35, and 40 C and fungal growth was measured daily. Linear regression analysis was performed on the data.

Red and mature green tomatoes of uniform color and size were selected and surface-sterilized to test the efficacy of fungicides. Each fruit was punctured 5 mm deep with a nail head in four numbered quadrants. Fruit were submerged in nutrient broth containing spores (10^5/ml) of *G. candidum* or *G. penicillatum*, air-dried for 1 hr, and dipped for 2.5 min in one of the following: nutrient broth, sodium hypochlorite (1 g/L), benomyl (0.5 g/L), imazalil (1 g/L), thiophanate methyl (1.4 g/L), sodium bicarbonate (3 g/L), potassium sorbate (1–2 g/L), vinclozolin (1.2 g/L), ferbam (1 g/L), or CGA-64251 (Vangard) (1.5 g/L). Each treatment was applied to two replicates each with five red and five mature green fruits. Ten fruits of each color were used as uninoculated, untreated checks.

Dipped fruit were air-dried and placed on fiber trays covered with perforated plastic bags. All treatments were stored at 20 C and lesion diameter was measured at 3 and 5 days.

RESULTS

There was a high degree of variability in rate of lesion development by the two fungi on red tomatoes although *G. candidum* appeared to invade fruit slightly faster than *G. penicillatum* (Table 1). Maximal growth of both fungi was observed at 25 C and minimal growth occurred at 5 C. *G. penicillatum* formed lesions significantly slower than *G. candidum* on mature green fruit at all temperatures studied. Growth of both fungi was significantly inhibited on green fruit compared with red fruit (Table 1). A distinctive sour odor accompanied decay of fruit inoculated with *G. candidum* but was absent in fruit inoculated with *G. penicillatum*.

In a second series of experiments, the growth responses of the two fungi were compared on PDA (Table 2). The growth responses of the two fungi to temperature were similar (both grew at 5–35 C); however, the growth rate of *G. penicillatum* was significantly slower than that of *G. candidum* at 15, 20, 25, and 30 C. Maximal growth of both fungi was observed at 30 C and minimal growth occurred at 5 C. Growth was not visible on any PDA plates incubated at 40 C.

Differences in growth characteristics of the two fungi could be discerned in culture. *G. penicillatum* produced more aerial hyphae than *G. candidum*, producing a fluffy texture; *G. candidum* grew appressed to the agar and appeared glossy, as though wet. Both fungi were compared with cultures on file at the ATCC in Rockville, MD.

In a third set of experiments, the response of inoculated fruit to fungicide treatments was evaluated. None of the fungicides prevented decay caused by the two fungi but they did affect rate of lesion development. Unprotected fruit were decayed at a rate not significantly different from that indicated in Table 1 at 20 C. Most fungicide treatments enhanced lesion development. Treatments may have injured the fruit, enhancing colonization by the fungi (Fig. 1). Sodium hypochlorite reduced the size of lesions formed by *G. penicillatum* on mature green and red fruit; however, it stimulated lesion formulation by *G. candidum*. Benomyl slightly inhibited lesions of *G. penicillatum* on green fruit, whereas imazalil slightly inhibited those

Table 1. Comparison of growth rates of *Geotrichum candidum* and *G. penicillatum* on red and green tomatoes at various temperatures[a]

		G. candidum		*G. penicillatum*	
Fruit maturity	Temperature (C)	Growth rate (mm^2/day)	R^2	Growth rate (mm^2/day)	R^2
Red	5	0.06	0.68[b]	0.04	0.77
	10	0.88	0.89	0.41	0.86
	15	1.63	0.79	1.58	0.81
	20	7.74	0.84	6.88	0.89
	25	11.22	0.85	10.41	0.64
Mature green	5	0.03	0.84	0.02	0.89
	10	0.13	0.98	0.08	0.86
	15	0.26	0.99	0.14	0.95
	20	0.55	0.98	0.25	0.99
	25	0.85	0.89	0.32	0.93

[a] Growth expressed as increase in lesion area.
[b] Coefficient of variability.

Table 2. Comparison of growth rates of *Geotrichum candidum* and *G. penicillatum* on potato-dextrose agar (PDA) at various temperatures[a]

	G. candidum		*G. penicillatum*	
Temperature (C)	Growth rate (mm^2/day)	R^2	Growth rate (mm^2/day)	R^2
5	0.45	0.97[b]	0.18	0.72
10	5.64	0.98	4.83	0.88
15	9.51	0.84	7.74	0.98
20	54.60	0.81	41.83	0.83
25	81.03	0.97	63.87	0.91
30	86.22	0.98	77.56	0.94
35	10.17	0.81	9.84	0.79
40	...[c]		...	

[a] Growth expressed as increase in colony area.
[b] Coefficient of variability.
[c] No growth observed.

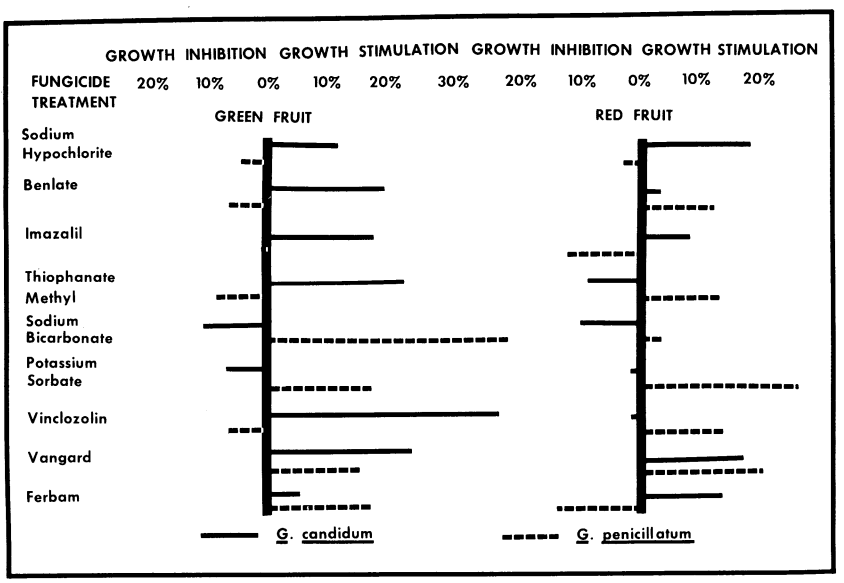

Fig. 1. Comparison of effects of nine fungicide treatments on rate of lesion development in red and mature green tomato fruit stored at 20 C. Percentage change in lesion diameter is in relation to inoculated, untreated fruit.

formed on red fruit. Thiophanate methyl reduced lesions of *G. penicillatum* slightly on green fruit and of *G. candidum* on red fruit. Sodium bicarbonate and potassium sorbate slightly inhibited lesions of *G. candidum* on mature green and red fruit. Vinclozolin only inhibited *G. penicillatum* slightly on green fruit. CGA-64251 had no inhibiting effect on either fungus, whereas ferbam reduced *G. candidum* growth only on red fruit.

DISCUSSION

Rots caused by *G. candidum* and *G. penicillatum* are very similar and can easily be confused. The most striking difference is that *G. candidum* produces a sour odor on decaying fruit, hence the name "sour rot." Fruit decayed by *G. penicillatum* had no comparable odor and the skin of decayed fruit tended to split readily. These fungi were easily recovered from stored ripening fruit. Both have been recovered from soil (2,4) and attack fruit with broken or punctured skin. Both fungi are wound pathogens because they cannot penetrate the skin of healthy fruit (3,7,14,17). On damaged fruit, however, the fungi are aggressive pathogens and can be detected within 24–28 hr of inoculation. Rotted areas appeared as water-soaked lesions with rapidly growing fungal colonies developing; these extended from the stem to the blossom end of infected fruit. The optimal growth temperature for *G. candidum* was near 30 C; its maximal growth temperature reported in the literature is 38.5 C and the minimum is 2 C (6). The optimal growth temperature for *G. penicillatum* is also near 30 C and its maximal growth temperature is between 38 and 40 C (Table 2); therefore, it is difficult to separate the two species on the basis of temperature response.

G. candidum grew more rapidly than *G. penicillatum* at storage temperatures and on the three substrates. The high degree of variability of decay observed on red fruit (Table 1) may have been due to differences in maturity of those fruit, which although selected visually for uniformity, could have had considerable differences in physiological maturity (20). Comparison of growth rates on red and mature green fruit (Table 1) shows the significant impact of fruit maturity on decay development. Both fungi grew more rapidly on PDA plates than on tomato fruits. It appears that *G. candidum* may be a slightly more aggressive pathogen than *G. penicillatum*.

Fungicide treatments were not as successful as had been anticipated; however, some treatments may bear further testing. It is interesting that sodium bicarbonate retarded growth of *G. candidum* but stimulated growth of *G. penicillatum* (Fig. 1). On the other hand, imazalil, which was the most effective against *G. penicillatum*, was not effective against *G. candidum*. There was actually more stimulation than inhibition of growth by most treatments by day 5.

Ferbam was one of the most effective treatments for control of *G. penicillatum* growth. Sodium bicarbonate was the most effective in controlling *G. candidum*.

There is a difference in response of the two fungi to the chemicals evaluated (Fig. 1). Some of this effect may have been a response to change in pH at the infection site caused by some chemicals used in the tests, as evidenced by stimulation of *G. penicillatum* growth on green and red fruit in response to potassium sorbate and green fruit treated with sodium bicarbonate (Fig. 1). Strains of *G. candidum* have been shown to differ in their responses to pH (12).

Because the fungi are not of major importance as postharvest pathogens of tomato fruit, one may be tempted to minimize the differences between the two species and consider them one; however, differences in response to chemicals may be important for other fruits and vegetables. We are continuing to evaluate a number of *Geotrichum* isolates from several sources that may give us additional insight into the behavior of this group of postharvest pathogens.

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