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Analysis of the Relationship Between Temperature and Vine Declines Caused by *Acremonium cucurbitacearum* and *Monosporascus cannonballus* on Muskmelon

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ABSTRACT

Temperature plays a major role in the incidence and severity of many plant diseases. It has been proposed that temperature exerts considerable influence on the development of some muskmelon vine declines. *A. cucurbitacearum* had a growth optimum at 25°C and the disease is associated more with cooler climates as compared to *M. cannonballus*. *M. cannonballus* had a growth optimum of 30°C and may have a threshold soil temperature requirement for the development of severe vine decline. Mean daily maximum and minimum temperatures of ≥35/20°C near the end of the growing season appear to be associated with vine decline caused by *M. cannonballus*. In contrast, temperatures of <30/20°C near the end of the growing season appear to be more conducive to development of vine decline caused by *A. cucurbitacearum*. Preliminary evidence suggests that temperatures in melon production areas where *A. cucurbitacearum* and *M. cannonballus* are present may exert a great deal of influence on the development of vine decline.

RESUMEN

La temperatura desempeña un papel importante en la incidencia y la severidad de muchas enfermedades de las plantas. Se ha propuesto que la temperatura ejerce una influencia considerable en el desarrollo de algunas pudriciones de la enredadera del melón. *Acremonium cucurbitacearum* tuvo un grado óptimo de crecimiento a 25°C y la enfermedad se asoció más con climas más frescos en comparación con *Monosporascus cannonballus*. *M. cannonballus* tuvo un grado óptimo de crecimiento a 30°C y probablemente puede requerir un umbral de temperatura del suelo para desarrollar una pudrición severa de la enredadera del melón. Las temperaturas máximas y mínimas promedio diarias de 35/20°C cerca del final de la estación de crecimiento parecen estar asociadas con la pudrición de la enredadera causada por *M. cannonballus*. En contraste, las temperaturas de <30/20°C cerca del final de la estación de crecimiento parecen conducir más al desarrollo de la pudrición de la enredadera causada por *A. cucurbitacearum*. Las evidencias preliminares sugieren que las temperaturas en las áreas de producción de melón donde están presentes *A. cucurbitacearum* y *M. cannonballus* pueden ejercer mucha influencia sobre el desarrollo de la pudrición de la enredadera.

Additional index words: Cucumis melo, collapse, Cucurbitaceae, sudden wilt, vine decline, temperature

Melon response to soilborne disease is determined by host resistance, inoculum density, pathogen virulence, and environmental conditions. Susceptibility of melon roots to infection and/or disease development varies with age of the plant as well as with changing environmental factors (Bruton, 1998; Bruton et al., 1998). The plant root and pathogen develop and function in dynamic thermal environments that change diurnally and seasonally. While some pathogens are capable of causing disease over wide geographic areas where susceptible plants are grown, other pathogens are less tolerant of environmental variations and may be restricted in their geographic distribution. Temperature has been suggested to play a major role in the incidence and severity of muskmelon vine declines (Bruton, 1998; Kim et al., 1995; Krikun, 1985; Martyn and Miller, 1996; Pivonia et al., 1997; Reuveni et al., 1983; Stanghellini et al., 1995; Wolff, 1996).

Monosporascus spp. cause vine decline in hot arid to semi-arid environments Martyn and Miller (1996). Two species of *Monosporascus* have been reported causing vine decline of cucurbits. *M. eutypoides* (Petrak) von Arx was first reported on melons in Israel (Reuveni and Krikun, 1983). Mertely et al. (1991) subsequently demonstrated that vine decline of melons in the Lower Rio Grande Valley of Texas was caused by *M. cannonballus* Pollack & Uecker (Pollack and Uecker, 1974). Lobo-Ruano (1991) reported *M. cannonballus* causing vine decline of muskmelon and watermelon in Spain, although isolates from Spain were identified by IMI as *M. eutypoides* (J.García-Jiménez and A.

Alfaro-Garcia, unpublished data). Sivanesan (1991a;1991b) has noted that *M. cannonballus* and *M. eutypoides* are similar, if not identical, in all morphological characters except in the number of ascospores/ascus and their ability to germinate. He concluded that they should be considered conspecific. In addition, recent examination of many isolates from Israel (M.E. Stanghellini and R. Cohen, personal communication) and Spain (unpublished data) clearly showed that *M. cannonballus* is the dominant species present in those countries since *M. eutypoides* was never found. Based on the existing information, *M. cannonballus* and *M. eutypoides* should be considered synonymous with *M. cannonballus* taking precedence (Bruton, 1998).

While *M. cannonballus* is clearly the cause of the predominant vine decline in the Lower Rio Grande Valley of Texas, southern Arizona, and southern California (United States), as well as in several other countries (Martyn and Miller, 1996), there is some question as to its importance in Spain. Lobo-Ruano (1991) first reported *M. cannonballus* causing a vine decline in Spain, although, no pathogenicity studies were demonstrated. Preliminary reports of pathogenic and non-pathogenic isolates have been made from Spain, although critical information on the inoculation procedure and assessment of virulence is lacking (García-Jiménez et al., 1994; Lovic, et al., 1993, 1996). Consequently, there has been some controversy as to the role of *M. cannonballus* in vine decline of melons in Spain.

Another vine decline which has symptoms similar in many respects to Monosporascus vine decline (Martyn and Miller, 1996; Mertely et al., 1991) is Acremonium collapse (García-Jiménez et al., 1989; 1994b; Gubler, 1996), although there are subtle differences. In contrast to Monosporascus vine decline, Acremonium collapse has been associated with cooler environments than Monosporascus vine decline (Bruton, 1998). García-Jiménez et al. (1989a; 1994) first reported a collapse or sudden death (collapso or muerta subita) of melons in Spain caused by *Acremonium cucurbitacearum* A. Alfaro-Garcia, W. Gams, & J.García-Jiménez (Alfaro et al., 1996). Since 1990, muskmelon production in the area of Valencia, Spain has almost been abandoned due to Acremonium collapse. Gubler (1982) reported a hypocotyl rot of melons in California caused by *A. cucurbitacearum*, but did not associate the disease with a late-season vine decline. More recently, *A. cucurbitacearum* has been associated with a vine decline syndrome of melons in the upper San Joaquin and Sacramento Valleys of California (Bruton et al., 1995; Gwynne et al., 1997). In the Lower Rio Grande Valley of Texas, *A. cucurbitacearum* has been isolated from muskmelon and watermelon (Bruton et al., 1996). It is unclear why *A. cucurbitacearum* does not cause a detectable level of disease in the Lower Rio Grande Valley of Texas (Bruton, 1998). However, Texas and Spain isolates of *A. cucurbitacearum* appear to be similar in level of virulence (Bruton et al., 1996) using the method of virulence assessment developed by Popham et al. (1998).

McDonald (1994) noted that distinguishing whether the soil environment is acting negatively on the host plant or positively on the plant pathogen may be difficult to ascertain, but absolutely critical to understanding disease epidemiology. Thermal requirements may be one of the key factors determining the compatibility of species in different environments and thus their distributions (Trudgill, 1996). Information on the influence of temperature on incidence and severity of many cucurbit vine decline diseases is lacking. As a result, this study was undertaken to 1) determine temperature growth optima of *A. cucurbitacearum* and *M. cannonballus* isolates from Spain and Texas and 2) explore the relationship of temperature on occurrence of vine decline of melons caused by *A. cucurbitacearum* and *M. cannonballus*.

MATERIAL AND METHODS

Fungal growth of Texas and Spain isolates under different temperatures. *A. cucurbitacearum* and *M. cannonballus* from Spain (3 isolates of each fungus) and Texas (3 isolates of each fungus) were arbitrarily selected to determine the optimum temperature for growth (Table 1). The respective fungi were previously isolated from muskmelon roots exhibiting symptoms of vine decline. Isolates stored in glass vials containing sterilized artificial soil (Terra-Lite,

Table 1. Description of fungal isolates used in temperature studies.

Fungus	Isolate #	*C. melo*/group	Location isolated	Date Isolated
Acremonium cucurbitacearum				
	SP 934941	*inodorus*	La Llosa (Castellón)	1993
	SP 875033	*inodorus*	Pueblo Nuevo (Valencia)	1987
	SP 934943	*inodorus*	El Romani (Valencia)	1993
	TX 951054	*cantaloupensis*	Starr County (Texas)	1995
	TX 941050	*cantaloupensis*	Hidalgo County (Texas)	1994
	TX 951065	*cantaloupensis*	Starr County (Texas)	1995
Monosporascus cannonballus				
	SP 935039	*inodorus*	El Romani (Valencia)	1993
	SP 935043	*inodorus*	El Romani (Valencia)	1993
	SP 935051	*inodorus*	La Llosa (Castellón)	1993
	TX 902020	*cantaloupensis*	Starr County (Texas)	1990
	TX 912035	*cantaloupensis*	Starr County (Texas)	1991
	TX 921999	*inodorus*	Starr County (Texas)	1992

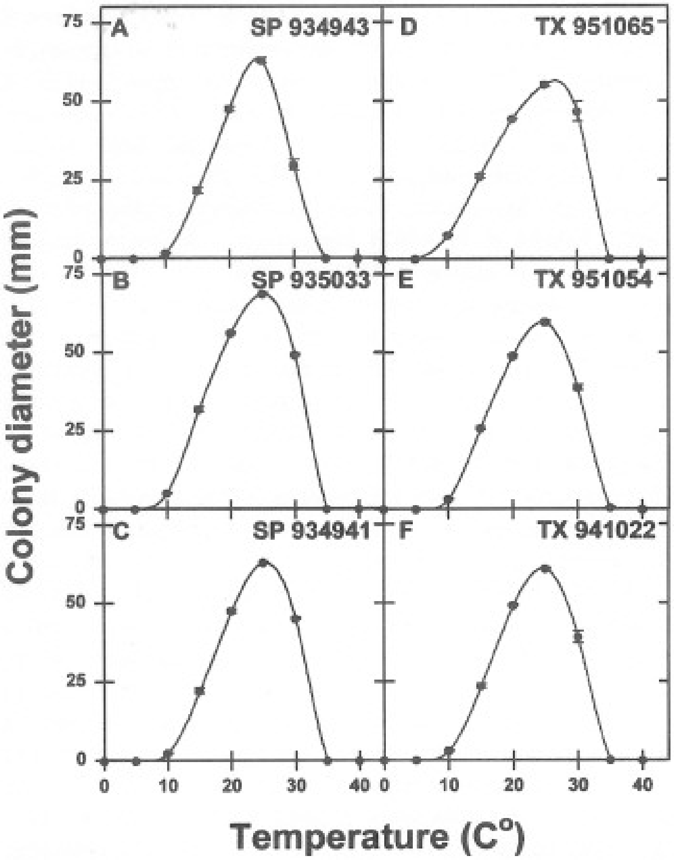

Fig. 1. Radial growth on PDA of Spain and Texas *A. cucurbitacearum* isolates in relation to temperature (13 days). Bars are standard errors of the mean.

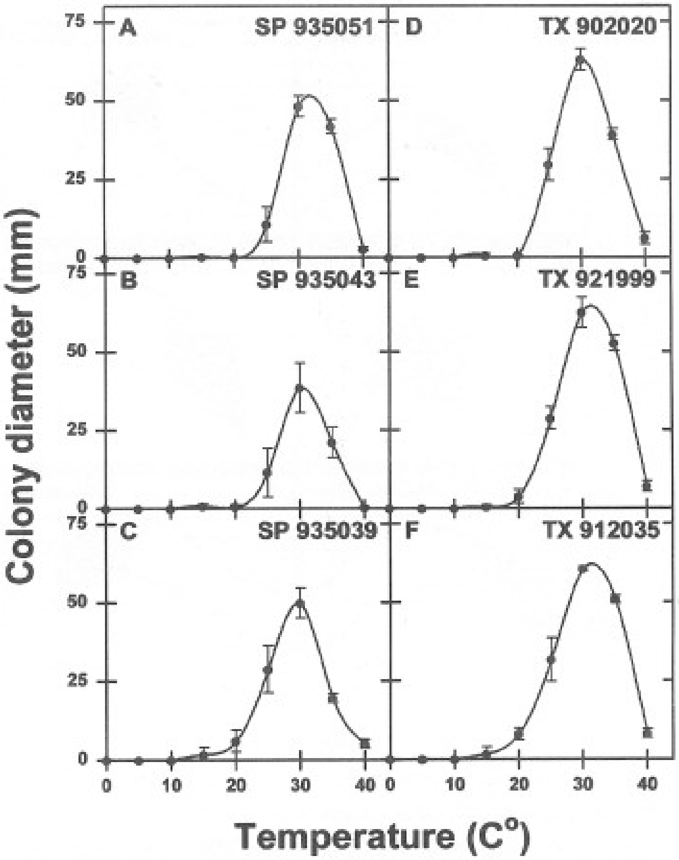

Fig. 2. Radial growth on PDA of Spain and Texas isolates of *M. cannonballus* in relation to temperature (3 days). Bars are standard errors of the mean.

Scotts-Sierra Hort. Products Co., Marysville, OH) were transferred to PDA and allowed to grow. After 7 and 4 days growth on PDA for *A. cucurbitacearum* and *M. cannonballus*, respectively, 7-mm PDA discs were taken from the outer perimeter of growth and transferred to the center of PDA plates. The plates (10 reps/trt) were incubated in environment chambers set at 5 to 40ºC at 5º increments. The diameter of fungal colonies was recorded at 13 and 3 da for *A. cucurbitacearum* and *M. cannonballus*, respectively. The experiments were repeated.

Temperature data from different melon growing areas. Temperature data were provided by J. K. Westbrook, USDA-ARS, College Station, TX. Data were obtained from the USDA, National Climatic Data Center, National Oceanic and Atmospheric Administration, Asheville, NC, for selected melon production areas where *A. cucurbitacearum* and/or *M. cannonballus* have been isolated. The mean maximum and mean minimum daily temperature was plotted for Sacramento (California), Rio Grande City (Texas), and Valencia (Spain) corresponding to areas where *A. cucurbitacearum* has been reported. Temperatures were also plotted for locations near to where *M. cannonballus* has been isolated i.e. Eilat (Israel), El Centro (California), and Phoenix (Arizona). Both fungi have been reported on cucurbits in Valencia and Rio Grande City. Temperature data for non-United States locations was averaged over a minimum of 13 yr and 30 yr for United States locations.

Statistical analysis. To determine the effect of temperature on fungal growth, ANOVA were done with model effects: isolate, temperature, and isolate by temperature interaction. Means comparisons utilized the Ryan-Einot-Gabriel-Welch Q Test at $P \leq 0.05$.

RESULTS

Fungal growth of Spain and Texas isolates under different temperatures. The analysis of variance showed no significant ($P \leq 0.05$) difference between Spain and Texas *A. cucurbitacearum* isolates with respect to optimum temperature for growth. *A. cucurbitacearum* grew best at 25ºC followed by 20, 30, 15, and 10ºC (Fig. 1). No growth occurred at 35ºC, regardless of isolate or origin.

The temperature for greatest ($P \leq 0.05$) growth of *M. cannonballus* isolates was 30ºC followed by 35 and 25ºC (Fig. 2). Little growth occurred at 15, 20, and 40ºC. However, there was a significant difference in growth of the various isolates. All Texas isolates grew significantly faster than Spain isolates at 30ºC.

Temperature data from different melon growing areas. Meteorological data are illustrated in Fig. 3 for melon production areas where *A. cucurbitacearum* and/or *M. cannonballus* have been previously isolated. The daily mean maximum and minimum temperature for Valencia, Spain are 20/11ºC at the beginning of the season and 28/20ºC at the end of the season. Temperatures in Sacramento, California are

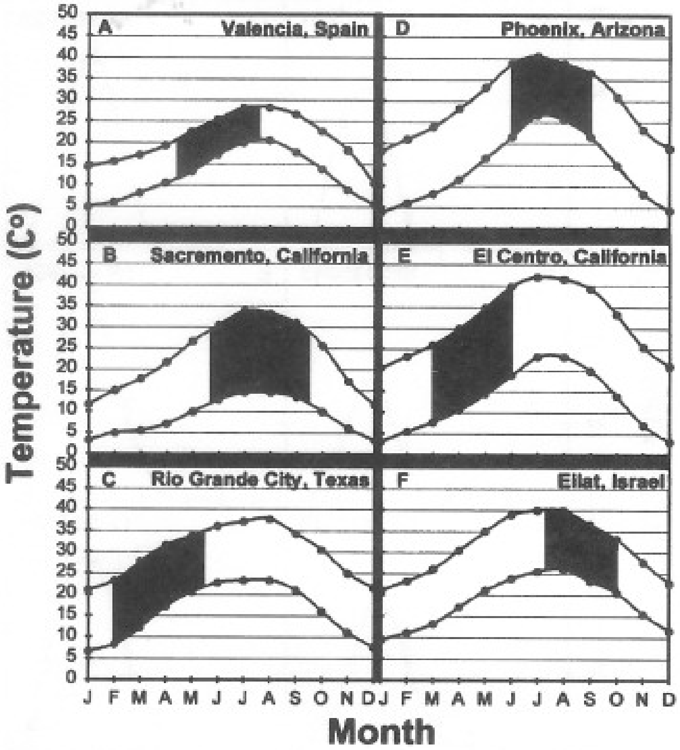

Fig. 3. Mean daily maximum and minimum temperatures for areas from which *A. cucurbitacearum* (A-C) and *M. cannonballus* (A, C-F) have been isolated. Shaded portion represents the primary melon growing months for each production area where vine declines have been prevalent.

considerably higher than in Valencia during the melon growing season, however, the mean minimum temperatures are much lower which undoubtedly moderate the soil temperatures. In contrast, temperatures at Rio Grande City, Texas are similar to those in Valencia at the beginning of the season, but the mean maximum temperature is ca. 7°C higher (35°C) in Rio Grande City at the end of the growing season. The mean maximum daily temperatures of Phoenix, El Centro, Rio Grande City, and Eilat (Arava region) where *M. cannonballus* occurs were ca. 35 to 40°C with a mean minimum of about 20°C at the end of the primary melon growing months.

DISCUSSION

Soil temperatures exert considerable influence on root diseases. Both the optimal temperature for growth and the range of temperatures that a fungus can tolerate determine whether it will survive and what role it will play in a given ecosystem. Although extrapolating "in vitro" temperature optima for growth of fungi to be the temperature optima for infection and/or disease development can be informative, it can also be misleading (Colhoun, 1973). There are several examples in which the optimum temperature for fungal growth differs dramatically from the optimum temperature for disease development (Campbell and Neher, 1994; Colhoun, 1973). However, there are many examples where the optimum temperature for fungal growth correlates with disease severity.

A. cucurbitacearum had optimum growth at approximately 25°C with a range of 10 to > 30°C. No growth occurred at 35°C. These values are similar to those reported by Armengol (1997) for Spanish *A. cucurbitacearum* isolates. *A. cucurbitacearum* causes a severe vine decline of muskmelon in Spain (García-Jiménez et al., 1994) and has been isolated consistently from collapsed muskmelon vines in the upper San Joaquin and Sacramento Valleys of California (Bruton et al., 1995; Gwynne et al., 1997). Temperatures during the primary melon-growing season in Valencia, Spain are relatively cool with a mean daily maximum/minimum temperature of 20/11°C at the beginning of the season to 28/20°C at the end of the season. The temperatures during the melon growing season in the Sacramento Valley are somewhat different with a mean daily maximum/minimum temperatures at the beginning of the season of 28/11°C and 32/13°C at the end of the season. However, the low night temperature in Sacramento no doubt moderates the effects of the higher day temperatures. Although *A. cucurbitacearum* has been isolated from melons in the Lower Rio Grande Valley of Texas (Bruton et al., 1996), the fungus does not cause a detectable level of damage (Bruton and Miller, unpublished data). Both day and night temperatures are higher toward the end of the growing season in the Lower Rio Grande Valley of Texas as compared to those in Valencia and Sacramento. In Rio Grande City, Texas, the mean daily maximum/minimum temperatures at the beginning of the season are 23/8°C and 35/22°C at the end of the season. Some Spain and Texas isolates are genetically very similar since they share the same vegetative compatibility grouping (Abad et al., 1997; Vicente et al., 1996). This would imply that temperature may play a greater role in vine decline severity caused by *A. cucurbitacearum* than genetic differences in the fungus.

M. cannonballus isolates from both Spain and Texas exhibited optimum growth at 30°C with a range of <20 to >40°C which is consistent with *M. cannonballus* isolates from Israel (Reuveni and Krikun, 1983) and Japan (Watanabe, 1979). This is in contrast to four Texas *M. cannonballus* isolates reported by Martyn and Miller (1996) in which equal growth occurred from 25 to 35°C and one isolate had equivalent growth over the range of 20 to 35°C. Vine decline, caused by *M. cannonballus*, has generally been associated with hot arid to semi-arid environments (Martyn and Miller, 1996). However, the relationship between soil temperature and disease incidence has not been delineated. Reports of *M. cannonballus* causing a vine decline, quick decline, collapse, or sudden wilt of cucurbits, has been reported from Guatemala (Bruton and Miller, 1997b), Honduras (Bruton and Miller, 1997a), Mexico (Martyn et al., 1997), and Saudi Arabia (Karlatti et al., 1997) in addition to those cited by Martyn and Miller (1996). The fungus has been found indigenous to desert areas in Arizona (Stanghellini et al., 1996). However, it is not always clear from the literature as to what environmental conditions the melon crops are subjected to during the growing season because of the use of plastic mulch, plastic tunnels, topographical data, and air/soil temperatures. The temperatures in the Jordan Valley and Arava region of Israel during the summer range from 23°C at night to 38°C in the day (Reuveni and Krikun, 1983) which correspond closely with the temperatures illustrated for Eilat in the present study. Reuveni et al. (1983)

demonstrated that soil temperatures of 30/20°C were considerably more conducive to disease development by *M. cannonballus* than temperatures of 25/20° C. Although Cohen et al. (1996) did not identify the causal organism, they noted that melons grown in late summer (high temperature) may be totally destroyed by sudden wilt in Israel. However, spring crops in the same fields may be unaffected. Pivonia et al. (1997) later determined that *M. cannonballus* was the major cause of sudden wilt in the Arava region of Israel. Wolf (1996) reported similar results at Weslaco, Texas in which spring transplanted (27 March) cantaloupe had severe vine decline caused by *M. cannonballus*. In contrast, a fall transplanted (30 August) cantaloupe crop had a low incidence of disease. The mean daily maximum/minimum temperature of the fall transplanted crop reported by Wolf (1996) was 31/19°C at the beginning of the season and 26/14°C at the end of the season (data not shown). The mean daily maximum/minimum temperature for Phoenix, Arizona is 39/22°C at the beginning of the season to 37/22°C at the end of the season. A few degrees difference in temperature may be critical for severe vine decline caused *M. cannonballus*. Early root infection and rapid symptom development were associated with soil temperatures greater than 25°C at 10 cm soil depth (Kim et al., 1995; Krikun, 1985; Stanghellini et al., 1995). High night temperatures may be equally or more important to the development of Monosporascus vine decline as compared to high daytime temperatures (M. E. Stanghellini, personal communication). Although *M. cannonballus* is isolated from melons in several Spanish Provinces, temperatures in many of the areas including Valencia may be below optimum for development of vine decline caused by *M. cannonballus*. While *A. cucurbitacearum* appears to be the primary cause of vine decline in Spain, *M. cannonballus* does cause some damage (personal observations, Lobo-Ruano, 1991). In fact both fungi coexist in many fields (unpublished data). Pivonia et al. (1997) demonstrated that a combination of pathogens had a synergistic effect on disease severity.

In this study, we have attempted to show the relationship between temperature and the incidence of vine decline caused by *A. cucurbitacearum* and *M. cannonballus*. Mean daily maximum and minimum temperatures of $\geq$35/20°C near the end of the growing season appear to be associated with vine decline caused by *M. cannonballus*. In contrast, daily maximum and minimum temperatures of <30/20°C near the end of the growing season appear to be more conducive to development of vine decline caused by *A. cucurbitacearum*. Additional research on actual soil temperatures and disease incidence is needed to more accurately define the role and interactive effects of temperature on vine decline caused by *A. cucurbitacearum* and *M. cannonballus*.

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