Impact of *Monosporascus cannonballus* on root growth of diverse melon varieties and their F1 progeny in the field

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ABSTRACT

Genetic variation for resistance to *Monosporascus cannonballus* in melon has been demonstrated in several greenhouse experiments. The evaluation of eight diverse melon lines and F1 progeny for resistance to this pathogen under field conditions was carried out over two successive seasons in South Texas. Roots were excavated and rated on a 1 to 5 scale with 1 being disease free and 5 being dead and withered. Lateral root numbers were counted and narrow-sense heritability (h^2) estimates for all traits were calculated. 'Deltex' and its progeny exhibited low damage ratings corresponding well with greenhouse results. 'PI 403994' performed much better in the field than in the greenhouse tests and 'Créme de Menthe' x 'Caravelle' performed much poorer. Heritabilities for the inverse of the root damage rating were high in both seasons as was the h^2 for lateral root number. Additive genetic variation for these traits appears to be important. Additionally, there was a strong negative correlation (r = -0.71) between lateral root number and root damage rating. Improvement of both root structure and *M. cannonballus* resistance should be possible in a reasonable timeframe.

RESUMEN

La variación genética en la resistencia del melón a *Monosporascus cannonballus* ha sido demostrada en varios experimentos de invernadero. Se evaluó la resistencia a este patógeno de ocho diferentes líneas de melón y de la progenie F1 bajo condiciones de campo durante dos estaciones sucesivas en el sur de Texas. Las raíces fueron desenterradas y clasificadas en una escala de 1 a 5 donde 1 correspondió a libre de la enfermedad y 5 a muerto y marchito. Se contaron las raíces laterales y se calcularon las estimaciones de la capacidad hereditaria en sentido estrecho (h^2) para todas las características. 'Deltex' y su progenie exhibieron un grado de daño bajo lo cual concordó con los resultados de invernadero. 'PI 403994' presentó mejor desempeño en el campo que en las pruebas de invernadero y 'Creme de Menthe' x 'Caravelle' presentó peor desempeño. Las capacidades hereditarias para el inverso del grado de daño de la raíz fueron altas en ambas estaciones así como también lo fue el h^2 para el número de raíces laterales. La variación genética aditiva para estos rasgos parece ser importante. Además, se presentó una fuerte correlación negativa (r = -0.71) entre el número de raíces laterales y el grado de daño de la raíz. El mejoramiento de la estructura de la raíz así como de la resistencia a *M. cannonballus* debe ser posible en un lapso de tiempo razonable.

Additional index words: narrow-sense heritability, vine decline, germplasm

The decline of melon vines prior to fruit maturity is a growing problem in South Texas and other hot, arid regions (Cohen et. al., 2000). The destruction of roots by a complex of soilborne root pathogens reduces the plant's capacity to uptake water and nutrients from the soil, causing eventual collapse of the vine. *Monosporascus cannonballus* is the pathogen causing the most severe root destruction and vine decline in South Texas (Martyn and Miller, 1996). Several experiments in the greenhouse to evaluate the impact of this fungus on melon root and vine growth have been carried out (Mertely et al., 1991 and 1993b; Crosby et al., 2000). Screening diverse melon germplasm and F1 progeny revealed the existence of resistance and the likelihood that quantitative inheritance is involved (Crosby, 2000).

The severe inoculation conditions utilized in these screening tests provided good evidence that the resistance observed in several melon lines is genetic and heritable in nature. The usefulness of this resistance under field conditions requires more extensive screening in several locations with *M. cannonballus* infested soil.

The performance of a crop in a greenhouse is not always indicative of its field performance. The latter is much more important to a commercial producer despite the limitations for certain types of analyses. The evaluation of complex root traits is difficult under field conditions. However, several root and vine characteristics can be measured in the field. These can be related to *M. cannonballus* resistance as well as other horticulturally important traits. The goal of this research was to determine if improving root structural development and *M. cannonballus* resistance was a realistic breeding objective to create superior lines for commercial use. The following hypotheses were tested: superior root development in the greenhouse reflects superior root development in the field; resistance to *M. cannonballus* under controlled inoculation is sufficient under field conditions; and genotypes with superior root phenotypes and *M. cannonballus* resistance were adapted to other stresses in the field and possessed moderately good horticultural quality characteristics.

MATERIALS AND METHODS

Eight of the parents utilized in greenhouse experiments (Crosby, 2000) and seven individual crosses were selected to represent an array of genotypes. These included: ‘Deltex,’ ‘Doublon,’ ‘PI 124111 x TDI,’ ‘Creme de Menthe,’ ‘PI 403994,’ ‘Perlita,’ ‘Magnum,’ and ‘Caravelle.’ The first three possessed the highest levels of resistance to *M. cannonballus*, while the fourth exhibited moderate resistance. The last four were extremely susceptible under greenhouse conditions (Crosby, 2000).

First Field Trial. Seed was planted in styrofoam speedling trays, in individual cells, in a commercial peat moss/vermiculite media (Sunshine Mix, SunGro, Bellevue, WA) on March 3, 1998. After three weeks seedlings of each line were transplanted to a field on the Texas A&M Research Center, Weslaco in a RCB design with four replications and 24 plants per replication. Soil was Hidalgo Fine Sandy loam, heavily infested with *M. cannonballus* (Mertely et.al., 1993a). The seedlings were planted 60 cm apart in 100 cm beds with black plastic mulch and drip irrigation to emulate commercial production conditions. Plants were fertilized one week after planting with 109 kg N/ha in the form of urea. Prior to evaluations for vine decline, most plants became severely debilitated by powdery mildew. However, root ratings for *M. cannonballus* damage were made for each plant. The system for rating roots was a scale of 1 to 5: 1=no symptoms, 2=few tan lesions, 3=extensive tan lesions and necrosis of smallest roots, 4=extensive tan lesions, perithecia, necrosis of small and medium roots, 5=completely withered, necrotic, dead roots (Table 1).

Second Field Trial. A second experiment was conducted beginning in August, 1998. Seven of the above parents and six different crosses were planted. Procedures were the same as above except for the implementation of a spray program to control powdery mildew. The fungicide Myclobutanil (Nova, Rohm and Haas Co.) was applied at a rate of 174 g (a.i.)/ha. All plants were evaluated for vine decline and fruit quality. In addition to root ratings for *M. cannonballus* damage, numbers of lateral roots were counted for each plant. Root samples from each plant were collected in plastic bags and stored at 4ºC.

Isolation of *M. cannonballus* was attempted for each root sample. Under a laminar flow hood, four samples per root were cut into two cm pieces and surfaced sterilized by washing in a 10% bleach solution (5.25% NaOCL) for one minute. The pieces were rinsed with sterile distilled water for another minute and plated onto water agar. After three days in the laminar flow hood at 25ºC, mycelial growth from the root pieces was observed in 95% of the plates. A small disk of agar containing a mycelial tip was transferred from each plate to a fresh V8 agar plate (Mertely et al, 1991). After five days under

Table 1. Mean separations for root ratings and lateral root numbers of eight melon parents and seven progeny, heritabilities from parent-offspring regression, standard errors and correlation between Fall root rating and lateral root number, 1998.

	Spring Season	Fall Season	
Cultivar	Root rating[z]	Root rating	Number of laterals
Deltex x PI 403994	1.50 a[y]	1.75 ab	16 ab
PI 403994	1.88 ab	1.63 ab	13 bcde
Deltex	2.50 bc	1.38 a	18 a
Doublon	–	2.25 bcd	15 b
PI 124111xTDI x Caravelle	2.75 c	–	–
PI 124111xTDI	2.83 c	–	–
Deltex x Perlita	2.83 c	2.25 bcd	14 bc
Creme de Menthe	3.00 cd	2.75 bc	15 b
Magnum x Doublon	3.75 de	2.13 bc	15 b
Creme x Doublon	4.00 ef	2.75 cde	12 cde
Magnum	4.13 ef	3.50 fg	14 bc
Deltex x Caravelle	–	2.88 def	13 bcde
Caravelle	4.33 efg	3.88 gh	11 de
Perlita	4.63 fg	3.25 efg	10 e
Creme x Caravelle	5.00 g	4.25 h	13 bcde
h2	1.51	0.78	0.69
SE	1.71	0.24	0.62
Correlation with number of laterals	–	-0.71	1.0

[z]Root damage rating: 1=no symptoms, 2=few tan lesions, 3=extensive tan lesions and necrosis of smallest roots, 4=extensive tan lesions, perithecia, necrosis of small and medium roots, 5=completely withered, necrotic, dead roots.

[y]Mean separations by LSD, P≤0.05. Means with the same letter are not significantly different.

the flow hood at 25ºC, all plates were visually evaluated to determine the presence of *M. cannonballus*. Mean separations by LSD were calculated with StatGraphics Plus (Manugistics, Rockville, MD). Means and regression of offspring values onto mid-parent values were calculated using the data analysis tools of Microsoft Excel. Because increasing root ratings represented increased disease damage, the inverses of these values were utilized to allow the regression curve to predict the reduction of the disease damage. Heritabilities were calculated from the coefficient of the regression, b. Correlation analysis of the two traits was completed as well.

RESULTS

Comparison of root ratings revealed substantial variability among the parents and progeny. The root ratings were higher during the Spring, as disease damage was greater (Table 1). The higher temperatures and drier weather in addition to stress from powdery mildew were likely responsible. During the Spring, 'PI 403994' exhibited the least damage of any parent (1.88). This was significantly lower than everything except 'Deltex' (2.50). The cross of these two parents exhibited significantly lower root damage (1.50) than all other entries except 'PI 403994.' 'Perlita' (4.63) and 'Caravelle' (4.33) had the highest damage ratings among the parents. The cross of 'Creme de Menthe' and 'Caravelle' (5.00) had significantly more damage than all other lines except these two parents.

Lower damage ratings in the Fall could be attributed to cooler, wetter weather and control of powdery mildew. Once again, 'Deltex' (1.38), 'PI 403994' (1.63) and their F1 progeny (1.75) had the lowest damage ratings. 'Deltex' was significantly lower than everything but the latter two lines. 'Doublon' (2.25) and 'Creme de Menthe' (2.75) also showed limited damage. The progeny of 'Deltex' x 'PI 403994' were extremely uniform and vigorous with the largest root systems of any cross or parent. Fruit was small and sour but edible. The cross of 'Deltex' and 'Perlita' (2.25) exhibited significantly less root cross of 'Magnum' and 'Doublon' (2.13) also had significantly

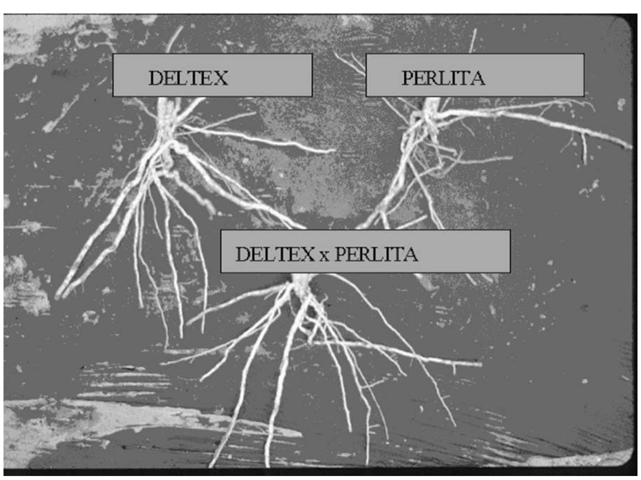

Fig. 1. Tolerant (Deltex) and susceptible (Perlita) melon (*Cucumis melo*. L) parents and their F_1 progeny grown in *Monosporascus cannonballus* infested soil.

damage than the susceptible parent, 'Perlita' (3.25, Fig. 1). The less root damage than the susceptible parent, 'Magnum' (3.50). Both of these crosses produced high quality fruit in most of the progeny. The poorest performer was again the cross of 'Creme de Menthe' and 'Caravelle,' (4.25) which had significantly more root damage than everything except 'Caravelle.'

Heritabilities from parent-offspring regression were extremely high for reduced root damage and moderately high for number of laterals (Table 1). In addition, there was a negative correlation (-0.71) between number of laterals and the root damage rating.

M. cannonballus was isolated from more than 90% of the root samples from these melons. In addition several other melon root pathogens were isolated, including *Macrophomina phaseolina, Myrothecium roridum and Rhyzopicnis spp.*

DISCUSSION

The results complemented previous work by Wolff and Miller (1998) with regards to parent performance. 'Deltex' and 'PI 403994' had exhibited good tolerance to *M. cannonballus* in these previous field evaluations, while 'Caravelle' and 'Magnum' had not (Wolff, 1996). The progeny performance was important for several reasons. The high heritabilities for reduced disease damage in crosses involving tolerant parents was a positive discovery. The relatively small sample size may have inflated these values, as may have heterosis for specific crosses. Further investigations with more crosses would provide greater support for these results. Additive genetic variation appears to contribute to the resistance mechanism in the field as it did in the greenhouse (Crosby, 2000). Most progeny performed within the range of the parents as in the top section of Figure 1. However, heterosis may have also contributed to both disease resistance and root development and may have boosted the h^2 values. The F1 progeny of 'Deltex' x 'Perlita,'in the bottom section of Figure 1, demonstrates this point. The vigor of the cross between 'Deltex' and 'PI 403994' also appeared to evince heterosis.

The relatively high heritability for lateral root number provides further support for additive gene control in root development. Additive variation was also determined to be important for lateral root number in Alfalfa. (Pederson et al., 1984). The development of more laterals allows the plant to produce a larger root structure to recover more water and nutrients from the soil. The relatively high positive correlation between this trait and reduced root damage suggests a potential role of lateral root development in the resistance to *M. cannonballus*. Cohen et. al. (2000) demonstrated that reduced frequency of irrigations stimulated larger root systems and reduced wilt due to *M. cannonballus*. It is not clear whether increased lateral development can be stimulated by stresses such as infection by *M. cannonballus*. It has been demonstrated that lateral root initiation is stimulated by some factor which moves from mature tissue to the region of lateral primordia (Torrey and Clarkson, 1975). This seems to be the case with the extensive formation of laterals near the crown of the melon plant where tissue is more mature. In other investigations suppression of lateral formation has been linked to taproot development within a certain distance from the apex (Torrey and Clarkson, 1975). All of these factors which influence lateral development are under varying degrees of genetic control. The results from this study suggest that there may be substantial genetic variation for these mechanisms leading to the phenotypic variation. The fact that this variation appears to be heritable in an additive fashion suggests that lateral root development may be improved in a predictable fashion by careful selection of parents. Development of lines with greater lateral production may be an important step towards reducing damage from *M. cannonballus*.

The performances of ‘PI 403994’ and the cross of ‘Creme de Menthe’ and ‘Caravelle’ contradicted results from greenhouse experiments (Crosby, 2000). The ‘PI 403994’ was extremely susceptible to *M. cannonballus* under high inoculation in sand culture. In contrast, this line grew extremely well in the field and exhibited few symptoms of *M. cannonballus* infection. This could be due to lower levels of the pathogen in the field soil or the interaction of the fungus with other soil micro-organisms. Whatever the reason, the drastic difference in response to the field environment suggests that this line may have an extremely variable range of phenotypic expression. Segregation for resistance could be possible but replication should have identified this as the reason. The responses were consistent over several progeny.

The poor field performance of cross ‘Creme de Menthe’ x ‘Caravelle’ did not seem unusual as one of the parents is highly susceptible to *M. cannonballus*. However, this cross performed well under inoculation in the greenhouse. This suggests the possibility of other factors in the field which may have limited the performance of this cross. Susceptibility to other diseases could have caused its decline. Several other melon pathogens besides *M. cannonballus* were isolated from root samples. Alternatively, the additional stresses of heat and disease pressure can increase susceptibility to *M. cannonballus* in the field (Cohen et. al., 2000). The value of field evaluations is brought to focus by these discoveries. Good performance under greenhouse conditions does not always imply good field performance. In addition, extremely high disease pressure under controlled inoculations may restrict performance of genotypes which grow well in the field.

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