Influence of Salinity and Root-knot Nematode as Stress Factors in Charcoal Rot of Melon

C. Nischwitz, M. Olsen and S. Rasmussen

Abstract

Incidence of Charcoal rot, caused by the soil borne fungus Macrophomina phaseolina, may be increased in some crops by the addition of stress on the host caused by high salinity of soil or irrigation water and infection by plant pathogenic nematodes. Since both of these factors may be problematic in melon production in Arizona, studies were initiated to determine if higher salt concentrations of irrigation water and infection by Root-knot nematode (Meloidogyne incognita) may be involved in recent increased incidences of Charcoal rot of melon. In greenhouse trials, higher concentrations of salts in irrigation water significantly increased the percentage of plants that died due to Charcoal rot. However, no significant difference was found in the percentage of dead plants inoculated with both root-knot nematode and M. phaseolina compared to plants inoculated with M. phaseolina alone. Results of these trials indicate that salinity may be a factor in the increased incidence of Charcoal rot of melon, but that root-knot nematode infection may not play a role.

Introduction

Charcoal rot of melon is caused by *Macrophomina phaseolina* (Tassi) Goid., a soilborne, microsclerotia producing fungus common in tropical and subtropical regions. *M. phaseolina* causes a root and stem rot on a large number of host plants including many important crops such as sorghum, sunflower, corn, melon and beans (Mihail, 1992). *M. phaseolina* generally infects many crops that are subject to severe stress caused by drought (Pande et al., 1989), high temperatures and flowering (Edmunds, 1964). Salinity and root-knot nematodes also can be major stress factors (El Mahjoub et al., 1979; Tu and Cheng, 1971; Siddiqui and Husain, 1991).

Charcoal rot causes crown decline and root rot in melon. *M. phaseolina* commonly infects the lower stem at the soil line causing water-soaked lesions that girdle the stem, but it also may cause root rot (Zitter, et al., 1996). It has been previously described as the most serious disease of melons in the Lower Rio Grande Valley of Texas (Bruton et al., 1985). As early as 1933, Charcoal rot was considered a serious problem (Taubenhaus and Ezekiel, 1933) and later was attributed to premature vine decline in cantaloupe in south Texas (Carter, 1979). Reuveni et al. (1982) reported collapse of drip-irrigated melon in Israel in both winter and summer conditions due to a root rot caused by *M. phaseolina*. More recently, vine decline of melon has been attributed to several soilborne pathogens including *M. phaseolina,* but pathogens included in the vine decline complex vary (Aegerter et al., 2000; Farr et al., 1998; Troutman and Matejka, 1970; Zitter, et al., 1996).

In the last few years, Charcoal Rot has become an increasingly important problem for melon growers in Arizona. Disease incidence has increased in drip-irrigated fields, whereas it has remained virtually unknown in furrow irrigated fields. It has appeared in drip-irrigated fields that have never been planted with melons, and has become increasingly widespread in fields with short rotations back to melons. Since disease is often associated with stress factors, it was puzzling why the disease appeared in drip-irrigated melons.

This is a part of the 2002 Vegetable Report, University of Arizona College of Agriculture and Life Sciences, index at: http://ag.arizona.edu/pubs/crops/az1292/

In drip-irrigated melons successively planted in the same beds for two or three crops, as many melons are now, salt can accumulate on the soil surface and at the margins of the wetted zone. Because salt distribution is moisture dependent (Hanks and Ashcroft, 1980), salts increase in beds that are not leached, especially at the soil surface, where *M. phaseolina* infections are most common. Another stress factor, Root-knot nematode, is a perpetual problem in melon production. It is a stress factor even at low levels of infestation since infections interfere with water and nutrient uptake and transport. In addition, plant pathogenic nematodes can lead to higher disease incidences by providing entrance sites for *Macrophomina phaseolina* (Tu and Cheng, 1971; Siddiqui and Husain, 1991).

The purpose of this study was to determine if increased salinity and Root-knot nematode infestations in soils contribute to increased incidence of Charcoal rot of melon. These were preliminary greenhouse trials to determine levels of increased salinity and nematode infestations that would be problematic in field soils in drip-irrigated melon production.

Materials and Methods

Greenhouse studies were established at the Campus Agricultural Center of The University of Arizona in fall 2000. A pasteurized soil:sand mixture (1:1) was air dried and infested with *M. phaseolina* by mixing *M. phaseolina* inoculum into the soil mix. The infested soil was stored in bins and used throughout this study. Inoculum consisted of colonized sorghum seed that was prepared in the laboratory two to four weeks previously. Sorghum seed was washed, soaked in tap water overnight and placed in Petri dishes, covering the bottom of the dish completely. Dishes were autoclaved twice for one hour on separate days. Agar plugs from potato dextrose agar cultures of *M. phaseolina* isolated from melon were used as inoculum, and plates were incubated at 30 C. After sorghum seed was completely colonized and masses of microsclerotia had formed within the sorghum seed substrate, the cultures were dried and blended to a coarse powder. The powdered inoculum was mixed into the sand to obtain a final inoculum density of about 100 microsclerotia per gram dry weight of soil (Mihail, 1992).

Irrigation water was amended to different salinities to determine the effect of salinity on disease. Since most fields in which disease has become a problem are drip-irrigated with CAP water, components of average CAP water were used for salinity adjustments (CAP web site http://www.cap-az.com/). Pre-determined ratios of NaCl, $MgCl_2$, and $Ca(NO_3)_2$ were used to adjust salinity levels from 0.53 dS/m in the tap water control to 1.16 dS/m (2x tap water), 3.1 dS/m (1.5x CAP water), 6.23 dS/m (4x CAP water) and 11.55 dS/m (6x CAP water). Preliminary studies determined that these salinities reduced growth but did not cause any other symptoms in plants grown in non-infested soil. All plants were irrigated daily to excess. In each of three trials, each treatment was replicated 10, 10 and 9 times. Each replicate was one pot with three to five plants per pot.

The effect of Root-knot nematode infection was determined by comparing the incidence of Charcoal rot in melon plants in soil infested with both *M. phaseolina* and Root-knot nematode to those in soil infested with *M. phaseolina* alone, Root-knot nematode alone and a non-infested control. Soil was infested with *M. phaseolina* as described above. *M. phaseolina* infested soils and non-infested soils were infested with different levels of Root-knot nematode by adding 1000 second stage juveniles (J2), 5000 J2 and 10,000 J2 to pots. Pots were irrigated with tap water.

In all trials, 'Greenflesh' honeydew melon (*Cucumis melo*) was direct seeded into infested soils in 10-cm pots in the greenhouse. Pots were placed in a greenhouse at 25˚C under a sodium-vapor light and watered daily. Plants were evaluated 24 days after the first symptoms of Charcoal rot appeared by counting numbers of dying and dead plants. To make sure plants had died from Charcoal rot, infection with *M. phaseolina* was determined by isolation from lower stems in all experiments. Numbers of infected dying or dead plants were used to calculate the percentage of plants that died from Charcoal rot or Root-knot nematode. Systat 9 was used to perform Analysis of Variance (ANOVA) to determine a treatment effect.

Results and Discussion

Increasing salinity of irrigation water increased the disease incidences of Charcoal rot significantly ($p < 0.001$) in all three trials (Table 1). The first symptoms of *M. phaseolina* appeared one week after irrigation treatments began. Twenty four days after initial disease symptoms, an average of 94%, 77%, 37%, 18% and 3% of plants died in treatments with 11.55 dS/m 6.23 dS/m, 3.10 dS/m, 1.16 dS/m and 0.53dS/m salinity levels, respectively. CAP irrigation water averages less than 1.0 dS/m, but as nutrients are added in drip irrigation water and as soil salinity increases in drip-irrigated beds, the salinities to which the plants are subjected increase through the season and from one crop to the next. Non-inoculated control plants in these trials were smaller when irrigated at the highest salinity compared to tap water, but none of the control plants died. These results indicate that salinity alone does not kill plants, but Charcoal rot of melon is increased with increasing salinity.

Root-knot nematode infestation did not influence disease incidences in these trials, and there were no significant differences between any of the treatments (ANOVA, $p = 0.127$). When plants were evaluated 24 days after inoculation, 93.9% of melon plants in treatments with *M. phaseolina* only, 83.9% in *M. phaseolina* + 1000 J2, 100% in *M. phaseolina* + 5000 J2, and 93.9% in the *M. phaseolina* + 10,000 J2 treatments were dying or dead. No plants died from Root-knot infection alone, but those inoculated with 5000 J2 or 10,000 J2 were stunted. These results show that Root-knot nematode is unlikely to increase Charcoal rot of melon and substantiate earlier reports that Root-knot nematode is not involved with increased incidence of Charcoal rot in melon in the Rio Grande Valley (Bruton and Heald, 1987).

Results indicate that high salinity levels may be a factor in Charcoal rot development leading to higher disease incidences while root-knot nematode infection is not. This may explain the increased incidence of Charcoal rot in subsurface drip-irrigated melons where soil salinity increases near the soil surface and at the wetting front of the drip zone creating an environment conducive to disease development. In fields with successive multiple melon crops, it may be possible to manage Charcoal rot by leaching soils between crops to reduce salinity.

References

Aegerter, B. J., T.R. Gordon and R. M. Davis. 2000. Occurrence and pathogenicity of fungi associated with melon root rot and Vine decline in California. Plant Disease 84:224-230.

Bruton, B. D., T. K. Hartz and E.L. Cox. 1985. Vine decline in muskmelon as influenced by cultivar and planting date. Hort. Science 20(5):899-901.

Bruton, B. D. and C. M. Heald. 1987. Effect of *Macrophomina phaseolina* and nematodes on cantaloupe. Phytopathology 77:1713.

CAP web site. 2000. http://www.cap-az.com.

Carter, W. W. 1979. Importance of *Macrophomina phaseolina* in vine decline and fruit rot of cantaloupe in south Texas. Plant Disease Reporter 63:927-930.

Edmunds, L.K. 1964. Combined relation of plant maturity, temperature, and soil moisture to Charcoal stalk rot development in grain sorghum. Phytopathology 54:514-517.

El Mahjoub, M., A. Bouzaidi, A. Joufri, A.Hamrouni, and El Beji. 1979. Influence de la Salinité des Eaux d'irrigation sur la Sensibilité du Tournesol au *Macrophomina phaseoli* (Maubl.) Ashby. Annales de Phytopathologie 11: 61-67.

Farr, D.F., M.E. Miller, and B.D. Bruton. 1998. *Rhizopycnis vagum* gen. et sp. nov., a new Coelomycetous fungus from roots of melons and sugarcane. Mycologia 90: 290-296.

Hanks, R.J. and G.L. Ashcroft. 1980. Applied Soil Physics. Yaron, B., Rommer, D.F.R., Sabey, B.R., Thomas, G.W., Vaadia, Y. and VanVleck, L.D. (eds.). Springer-Verlag

Mihail, J.D. 1992. *Macrophomina phaseolina* In: Singleton, L.L., Mihail, J.D. and Rush, C.M. (eds.), Methods for Research on Soilborne Phytopathogenic Fungi. APS Press, St. Paul.

Pande, S., L.K. Mughogho, N. Seetharama, and R.I. Karunakar. 1989. Effects of nitrogen, plant density, moisture stress and artificial inoculation with *Macrophomina phaseolina* on Charcoal rot incidences in grain sorghum. Journal of Phytopathology 126: 343-352

Reuveni, R., J., A. Kirkun, Nachmias and E. Shlevin. 1982. The role of *Macrophomina phaseolina* in a collapse of melon plants in Israel. Phytoparasitica 10: 51-56.

Siddiqui, Z.A. and S.I. Husain. 1991. Interaction of *Meloidogyne incognita* race-3 and *Macrophomina phaseolina* in a root-rot disease complex of chickpea. Nematologia mediterranea 19: 237-239

Taubenhaus, J.J. and W.N. Ezekiel. 1933. A new Charcoal rot of cantaloupe. Tex. Agric. Exp. Stn. Annu. Rep. (1932)45:74-75.

Troutman, J.L. and J.C. Matejka. 1970. Three fungi associated with cantaloupe roots in Arizona. Phytopathology 60: 1317

Tu, C.C. and Y.H. Cheng.1971. Interaction of *Meloidogyne javanica* and *Macrophomina phaseoli* in Kenaf root rot. Journal of Nematology 3: 39-42

Zitter, T. A., D. L. Hopkins and C. E. Thomas (eds.). 1996. Compendium of Cucurbit Diseases. APS Press, St Paul.

Table 1. Mean percentage of plants that died from Charcoal rot at different levels of salinity of irrigation water 24 days after initial disease symptoms appeared.

Salinity levels of irrigation water (dS/m)	**Trial****		
	1*	2*	3*
0.53 (tap water)	6.1 a	2.6 a	0 a
1.16	34.1 a	19.0 ab	0 a
3.10	47.8 ab	63.6 bc	2.4 ab
6.23	70.7 bc	97.5 c	65.9 b
11.55	100 c	97.6 c	85.7 c
ANOVA	P < 0.001	P < 0.001	P < 0.001

*Numbers followed by different letters are significantly different within a trial using the Tukey test for differences of means.

** Results from three trials using 10, 10 and 9 pots (replications) with 3 to 5 plants per pot.