# CONTROL OF MELOIDOGYNE MARYLANDI ON BERMUDAGRASS

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## ABSTRACT

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*Meloidogyne marylandi* is widely distributed on turf in Texas and is frequently associated with poor turf appearance and growth. Suppression of population densities of *M. marylandi* on established Bermudagrass through application of a new formulation of 1,3-dichloropropene (Curfew), fenamiphos (Nemacur), or the biological agent *Paecilomyces lilacinus* (MeloCon WG) was tested in separate experiments. Application of the fumigant 1,3-dichloropropene effectively suppressed nematode population densities for 10 weeks with a slight improvement in turf color (P = 0.08). Neither *P. lilacinus* nor fenamiphos effectively suppressed nematode population densities nor improved turf appearance. Fenamiphos did reduce root galling (P = 0.05) in one of two experiments. Juveniles of *M. marylandi* encumbered with *Pasteuria* spp. endospores were observed during these studies and the efficacy of this biological control agent in established turf needs to be investigated.

Key words: 1,3-dichloropropene, Bermudagrass, biological control, Cynodon dactylon, Meloidogyne marylandi, nematode management, Paecilomyces lilacinus, Pasteuria spp., root-knot nematode, soil fumigation, turfgrass.

## RESUMEN

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*Meloidogyne marylandi* se ecuentra ampliamente distribuido en céspedes en Texas y se asocia frecuentemente con mal aspecto y pobre crecimiento del césped. En experimentos independientes, se evaluó la supresión de poblaciones de *M. marylandi* en céspedes establecidos mediante la aplicación de una nueva formulación de 1,3-dicloropropeno (Curfew), fenamifos (Nemacur), o del agente biológico *Paecilomyces lilacinus* (MeloCon WG). La aplicación del fumigante 1,3-dicloropropeno suprimió las poblaciones de manera efectiva durante 10 semanas y mejoró ligeramente el color del césped (P = 0.08). Fenamifos y *P. lilacinus* no suprimieron de manera efectiva las poblaciones del nematodo ni mejoraron la apariencia del césped. Fenamifos sí redujo el agallamiento en las raíces (P = 0.05) en uno de dos experimentos. Durante estos estudios, se observaron endosporas de *Pasteuria* spp. en los juveniles de *M. marylandi* y es necesario investigar la eficacia de este agente de control biológico en céspedes establecidos.

*Palabras clave*. 1,3-dicloropropeno, céspedes, control biológico, *Cynodon dactylon*, fumigación del suelo, manejo de nematodos, *Meloidogyne marylandi*, nematodo agallador, *Paecilomyces lilacinus*, *Pasteuria* spp., pasto Bermuda.

## INTRODUCTION

*Meloidogyne marylandi* has been identified as a parasite of turfgrasses in Texas. The nematode was detected parasitizing a Bermudagrass pasture in Erath county in 1995, zoysia grass (*Zoysia matrella*) in Dallas county in 2003, and from Bermudagrass in Brazos and Refugio counties in 2004. In each instance, nematode identification was based on morphology and morphometrics of second-stage juveniles and males, and perineal patterns from females (Jepson and Golden, 1987; Golden, 1989). Esterase and malate dehydrogenase isozyme phenotypes of the populations from Dallas and Brazos counties were identical to those reported by Oka et al. (2003) for M. marylandi. A report of Hypsoperine graminis from a sports field near Abilene, where the Bermudagrass turf was exhibiting symptoms of poor growth (Orr and Golden, 1966), may have also been M. marylandi. In addition to Texas, M. marylandi has been reported from Arkansas (Elmi et al., 2000), Maryland (Golden, 1989), Israel (Oka et al., 2003) and Japan (Araki, 1992).

The Brazos county infestation was from Bermudagrass putting greens of a golf course established about 6 months earlier and each of the 18 greens was infested in November 2004 with nematode population densities ranging from 1,500 to 20,000 eggs and J2/500 cm<sup>3</sup> soil. It was subsequently determined that the turf farm in Refugio county that supplied the sprigs of Bermudagrass used to establish the greens was infested with M. marylandi. The infested putting greens were characterized by a low density of turf with a chlorotic to brown color. Collectively, these observations suggest that M. marylandi is widely distributed on several grass species in Texas and that it contributes to poor turf growth and appearance.

Despite the widespread distribution of *M. marylandi* and its frequent association with turf species exhibiting various symptoms of decline and poor growth, there are few reports on management of this species. The objective to this study was to evaluate the efficacy of two products for suppression of established nematode populations in intensively managed Bermudagrass on

golf course putting greens. The products evaluated included a new formulation of 1,3-dichloropropene intended for application to established turf (Curfew, Dow AgroSciences, Indianapolis, IN) and a commercial formulation of the biological control agent *Paecilomyces lilacinus* (Melo-Con WG, Prophyta, Poel, Germany). Fenamiphos (Bayer CropScience, Research Triangle Park, NC) was included in two tests as the industry standard, however this product is no longer labeled for use on turf.

#### MATERIAL AND METHODS

The efficacy of 1,3-dichloropropene (1,3-D) was evaluated in May of 2005 on two infested putting greens of a private golf course in Brazos county, Texas. The fumigant was applied by a commercial applicator to two 2.5-m wide plots across each green; each treated plot varied from 10 to 15-m long. Injection shanks were spaced 10 cm apart in the treated plots. Similar sized nontreated plots were established immediately adjacent to the fumigated plots. The fumigant (11.9 kg a.i./ liter) was applied to a depth of 10 cm at 4.7 liters/ha. Immediately after application of the fumigant, 12 mm of water was applied using a sprinkler irrigation system. Golf greens were planted to 'Tifdwarf' Bermudagrass (Cynodon dactylon) and constructed to United States Golf Association standard specifications.

Nematode population densities were estimated immediately prior to fumigation and at 2, 4, and 10 wk after treatment. Twenty soil cores (each 2.5-cm-diam by 10 cm depth) were collected from each plot to make one composite sample per plot. Nematodes were extracted from the soil and root samples by elutriation (Byrd *et al.*, 1976) and centrifugation (Jenkins, 1964). Eggs were extracted using 1% NaOCl (Hussey and Barker, 1973) from the root fragments recovered during elutriation. To determine viability of eggs extracted from the soil and roots, a sample of ca 1,000 eggs from each sample was placed separately into hatching chambers constructed from  $1 \text{ cm-diam} \times 1 \text{ cm-length PVC pipe with a}$ 20 µm (pore size) plastic screen glued to one end of the pipe section. These hatching chambers were placed in 5-cm-diam Petri dishes with sufficient water to cover the screen, and incubated at 25°C for 5 d. Total number of J2 that migrated through the screen was counted to estimate percentage hatch. Turf response to treatment was based on a subjective turf color rating (1 = light green color to 9 = dark green)and a turf root quality rating (1 = poor to 9)= excellent based on color and root length) (Morris and Shearman, 2006).

Two experiments were conducted to test the efficacy of P. lilacinus for control of M. marylandi. Both experiments were conducted on Bermudagrass putting greens at the same golf course as the experiment with 1,3-D. The first experiment with P. lilacinus was initiated in July 2005 on the same greens used for the previous fumigation test. This experiment had four replications of four treatments; nontreated, fenamiphos at 9.8 g a.i/m<sup>2</sup>, and P. lilacinus at 2.4 g and 4.9 g of product/m<sup>2</sup>. The experiment was a randomized complete block design, and each plot was  $6 \text{ m} \times 1.2$ m. The second experiment was initiated in March 2006 on a different green at the same golf course and had six treatments with four replications of each treatment: a nontreated control; fenamiphos at 9.8 g a.i./m<sup>2</sup>; *P. lilacinus* at 2 g/m<sup>2</sup> with at least 1.27 cm irrigation within 24 hr; P. lilacinus at 2 g/m<sup>2</sup> plus surfactant at 1.8 ml/m<sup>2</sup> (Affinity, Becker Underwood, Ames, IA) with at least 1.27 cm irrigation within 24 hr; *P. lilacinus* at  $2g/m^2$  as a drench treatment in a delivery volume of 12 l/m<sup>2</sup>; P. lilacinus at 2 g/m<sup>2</sup> plus a surfactant at  $1.8 \text{ ml/m}^2$  in a drench treatment in a delivery volume of  $12 \text{ l/m}^2$ . A second application of each treatment was made in April 2006, 6 wk after the initial application. Soil and root samples for each of these two experiments were collected at 4, 16, and 24 wk after initial treatment and processed as described above to estimate the effect of treatments on nematode population densities. Turf growth response to treatments was evaluated as described for the fumigation experiment.

All data were subjected to analysis of variance using the SAS general linear model (SAS, Inc., Cary, NC). Mean separations, when appropriate, were by Tukey's method.

#### RESULTS

Plots fumigated with 1,3-D had lower population densities of J2 and viable eggs at 2, 4, and 10 weeks after treatment (P =0.05) than the adjacent nontreated plots (Fig. 1). A high number of eggs were present in all soil and root samples collected at 2 weeks after treatment. However, less than 1% of the eggs from fumigated plots were viable based on percentage hatch over a 5-d incubation period, whereas greater than 60% of the eggs extracted from nontreated plots hatched in the same period. Although the mean population density of M. marylandi was lower in treated plots at 10 wk after treatment than in nontreated plots, the mean density was no longer different from the initial population density immediately prior to treatment. Turf color ratings were unaffected by treatment at 2 and 4 weeks after treatment but at 10 wk after treatment the color rating for the 1,3-D treated plots was 7.0 compared to 5.75 in the nontreated plots (P = 0.08). Turf root quality was unaffected by treatment at 2, 4, and 10 wk after treatment.

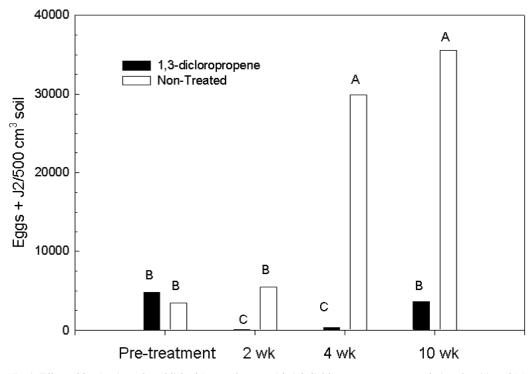


Fig. 1. Effect of fumigation of established Bermudagrass with 1,3-dichloropropene on population densities of *Meloidogyne marylandi*. Bars with the same letters are not different (P = 0.05).

In the first experiment to evaluate the efficacy of *P. lilacinus* for suppression of *M. marylandi*, no treatment effected a suppression of nematode population densities at any sample date (Fig. 2). Mean nematode densities were similar at 4 and 16 wk after treatment and then increased nearly 10-fold at 24 wk after treatment. Despite the absence of any effect of treatments on nematode population densities in the soil, fenamiphos treated plots had the lowest (P = 0.05) root gall rating at 24 wk after treatment (Fig. 3). No treatment had any effect on turf growth (data not shown).

In the second experiment to evaluate *P. lilacinus*, no treatment suppressed nematode population densities (Fig. 4). Similarly, there was no effect of treatment of on turf growth or color (data not shown).

## DISCUSSION

Plant-parasitic nematodes are often associated with poor turf growth. Of 442 turf samples from Texas submitted to different diagnostic laboratories in 1995, 10% were infested with Meloidogyne spp., with similar frequencies of Pratylenchus, Hoplolaimus, Trichodorus and Belonolaimus spp. (Stevens-Johnk, pers. comm.). Our recent observations indicate that M. marylandi is widely distributed on turf in Texas, and is often associated with poor turf quality. The Bermudagrass putting greens at the Brazos county site appeared to suffer the most damage from the nematodes following prolonged periods of irrigation during drought conditions. The local water source has a high concentration of

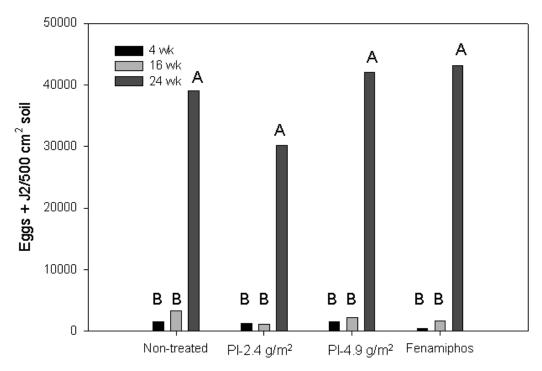


Fig. 2. Effect of treatment of Bermudagrass in the summer with a commercial formulation of *Paecilomyces lilacinus* and fenamiphos on population densities of *Meloidogyne marylandi*. Treatment effects were not significant but population densities at 24 wk after treatments were greater (P= 0.05) than those at earlier sample dates.

salts (ca 800 ppm Na) and it is possible that salt accumulation during these periods may be exacerbating the nematode damage. Turf quality appears to improve following several rainfall events that would be expected to leach accumulated salts from the root zone.

Efforts to suppress *M. marylandi* population densities met with mixed success. Fumigation with 1,3-dichloropropene was highly effective but had no residual activity and nematode populations densities quickly rebounded. Nonetheless, turf growth exhibited a positive response to the short-term suppression of the nematode populations. Additional research is needed to determine the long-term effects of multiple applications of this fumigant on both nematode populations and turf growth. Fenamiphos has been the industry standard for management of plant-parasitic nematodes on turf grasses, however in these

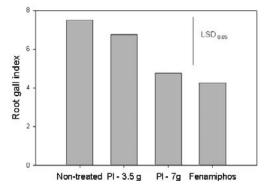


Fig. 3. Effect of treatment of Bermudagrass with a commercial formulation of *Paecilomyces lilacinus* and fenamiphos on root galling caused by *Meloidogyne marylandi* 24 weeks after treatment.

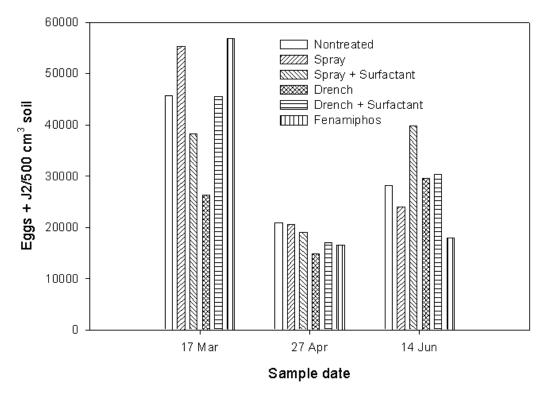


Fig. 4. Effect of treatment of Bermudagrass in the spring with a commercial formulation of *Paecilomyces lilacinus* as a soil drench or foliar spray, with and without a surfactant, and fenamiphos on population densities of *Meloidogyne marylandi*. All treatment effects were nonsignificant.

tests it had limited efficacy with regard to suppression of *M. marylandi* population densities. This material has been effective in improving Bermudagrass turf growth and color when parasitized by ectoparasitic species such as *Belonolaimus longicaudatus* and *Hoplolaimus galeatus* (Crow, 2005).

Paecilomyces lilacinus is a well known parasite of Meloidogyne spp. eggs (Chen and Dickson, 2004a) and it was hoped that the relatively shallow vertical distribution of *M. marylandi* on the putting greens and the coarsely textured soil would be favorable for the establishment and activity of this organism. Further, because nematode population densities were quite high in both experiments when the product was applied, it was hoped that the abundance of egg masses present would increase the probability of establishment of the fungus. Therefore, it was disappointing that the *P. lilacinus* product was not effective in these studies. Occasionally, J2 in these studies were observed to be encumbered with endospores of *Pasteuria* sp., the obligate bacterial parasite of numerous nematode species (Chen and Dickson, 2004b). This organism also has been reported to be effective in suppressing *Meloidogyne* population densities in many situations (Chen and Dickson, 2004b) and may be effective on *M. marylandi* on turfgrass.

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