# Factors Affecting Furfural as a Nematicide on Turf

J. E. Luc,<sup>1</sup> W. T. Crow<sup>2</sup>

Abstract: Recently a furfural nematicide Multiguard Protect EC was launched for use on turfgrasses in the United States. A series of greenhouse experiments were conducted to determine the concentration and exposure time required for this formulation to irreversibly affect *Belonolaimus longicaudatus*, and to study factors that might affect the practicality of furfural use in turfgrass systems. One experiment exposed *B. longicaudatus* to increasing concentrations of furfural (0 to 990 ppm) in vitro for either 24 or 48 hr, followed by inoculation onto bermudagrass. A second experiment evaluated effects of exposure of *B. longicaudatus* to increasing concentrations of furfural (9 to 990 ppm) in vitro for either 24 or 48 hr, followed by inoculation onto bermudagrass. A second experiment evaluated effects of exposure of *B. longicaudatus* to increasing concentrations of furfural an organic thatch layer. A third experiment evaluated effects on *B. longicaudatus* of increasing concentrations of *B. longicaudatus* to increasing numbers of *B. longicaudatus* with increasing furfural concentration beginning with the lowest concentration tested (270 ppm). *Belonolaimus longicaudatus* were virtually eliminated with furfural concentrations  $\geq 720$  ppm. Similarly, exposure to increasing concentration of furfural in soil solution in numbers of *B. longicaudatus*. Presence of thatch slightly reduced the population density of *B. longicaudatus*. Spray application of furfural only reduced numbers of *B. longicaudatus* at the two highest rates (3,600 and 4,950 ppm).

Key words: Agrostis palustris, Belonolaimus longicaudatus, bermudagrass, creeping bentgrass, Cynodon dactylon, furfural, nematicide, nematode management, sting nematode, turfgrass.

Plant-parasitic nematodes are viewed as one of the most important pathogen groups affecting turfgrasses (Crow, 2007). Among the most damaging and commonly found nematodes on turfgrasses in Florida is Belonolaimus longicaudatus (sting nematode) (Crow, 2005). Feeding by B. longicaudatus causes stunted rootgrowth, leading to decreased rates of water and nutrient uptake, and evapotranspiration (Johnson, 1970; Perry and Rhoades, 1982; Giblin-Davis et al., 1992; Busey et al., 1993; Luc et al., 2006). The recent removal of the organophosphate nematicide fenamiphos, which was the most commonly used nematicide on turfgrasses for decades, has increased the need for nematode management tools for turf. One of the more promising compounds investigated has been furfural (2furancarboxaldehyde), a naturally occurring aromatic aldehyde present in some essential oils, foods, and cosmetic products. Furfural is a contact nematicide and must be mechanically incorporated or moved into the soil profile with irrigation (Rodríguez-Kábana et al., 1993). Furfural is the active ingredient in several nematicides including CropGuard® and Protect®, which are being used in parts of Africa (Hensley and Burger, 2006). A nematicide containing 90% furfural, Multiguard Protect EC, was launched in the U.S. turfgrass market in 2011.

Furfural is degraded by bacteria and fungi in soil (Wierckx et al., 2011). Unpublished research performed by a manufacturer of furfural products (Illovo Sugar, Durban, South Africa) shows the half-life of furfural in soil to be 1.2 d under aerobic conditions and 1.8 d under anaerobic conditions (G. L. Burger, pers. com.). Because

it is rapidly degraded in soil, knowledge of the exposure time and concentration required to permanently affect nematodes is critical to determine its practicality as a turfgrass nematicide.

In turfgrass systems, thatch is an organic layer that forms on top of the soil that is composed of accumulated dead leaves, stems, rhizomes, and stolons (Smiley et al., 1992). Thatch has been shown to bind pesticides and to prevent movement of turf nematicides into the soil (Cisar and Snyder, 1996). The thickness of the thatch layer varies greatly with age, species, and cultivar of the turf, and with maintenance practices. Thicker thatch layers might interfere with the movement of furfural into soil and thereby reduce efficacy.

Turfgrass nematicides can be applied in different ways. Granular nematicides are typically applied using drop spreaders, fumigant nematicides are slit-injected into soil below the turf, and solutions and suspensions such as furfural are most often applied as a topical spray followed by irrigation to move the active ingredient into the soil. However, for small nematode affected areas "spot treatment" of nematicide may be applied as a drench in a high volume of water. Movement of furfural into soil is likely to differ with application method and amount of water used for application.

To better understand the utility of furfural as a nematicide, several lab and greenhouse experiments were conducted. The objectives of these experiments were to (i) quantify response of *B. longicaudatus* to increasing furfural concentration and exposure time, (ii) determine if presence of thatch interferes with furfural efficacy, and (iii) determine if surface spray application of furfural is effective against *B. longicaudatus* on turf.

#### MATERIALS AND METHODS

In 2011 several experiments were conducted at laboratory and greenhouse facilities of the University of Florida in Gainesville, FL. Turfgrass was grown in

Received for publication September 6, 2013.

<sup>&</sup>lt;sup>1</sup>Postdoctoral Research Associate, Entomology and Nematology Department, University of Florida, Gainesville, FL 32611.

<sup>&</sup>lt;sup>2</sup>Professor, Entomology and Nematology Department, University of Florida, Gainesville, FL 32611.

These studies were supported by contributions from Agriguard, LLC, and Illovo Sugar, LLC. E-mail: wtcr@ufl.edu

This paper was edited by Salliana R. Stetina.



FIG. 1. Effects of pre-inoculation exposure to increasing concentration of furfural and exposure time on subsequent *Belonolaimus long-icaudatus* population densities after 30 d on creeping bentgrass. Columns represent combined data from two identical trials, each with six replications. Columns with common letters are not different according to Fisher's LSD test ( $P \le 0.05$ ).

3.8-cm-diam.  $\times$  21-cm-deep UV-stabilized Ray Leach "Cone-tainers"™ (SC10, Stuewe & Sons, Inc., Tangent, OR) containing 100% USGA specification greens sand (USGA, 1993) with a 3-cm-deep plug of Poly-fil (Fairfield Processing, Danbury, CT) to prevent the sand from escaping from the drainage holes in the bottom. The total sand volume in each Cone-tainer was 130 cm<sup>3</sup>. B. longicaudatus inoculum was extracted using the decanting and sieving method (Cobb, 1918) from pure nematode cultures maintained on 'Penncross' creeping bentgrass (Agrostis palustris) in a greenhouse, and quantified by counting the nematodes in five 1-ml aliquots on a counting slide (Hawksley and Sons Limited, Lancing, Sussex, UK). Inoculum contained females, males, and juveniles, the percentages of each were not quantified. A single 1-cm-diam.  $\times$  2.5-cm-deep hole was made with a pencil into the soil at the surface of each Cone-tainer, into which the nematode inoculum was placed and the hole was closed. Greenhouses were maintained between 18 and 32°C. Furfural used in these experiments was the commercial 90% a.i. Multiguard Protect EC (Agriguard, Cranford, NJ) formulation supplied by the manufacturer. Statistical analysis was performed using SAS software (SAS Institute, Cary, NC). Each experiment was conducted in two separate trials, in all cases there were no differences among trials (P > 0.1), so data from both trials were combined for analysis.

Duration and rate study: The objective of this experiment was to quantify effects of increasing furfural solution concentration and exposure time on B. longicaudatus. Nematode-free aerial stolons of 'Champion' bermudagrass (Cynodon dactylon  $\times$  C. transvaalensis) were planted into Cone-tainers and allowed to produce roots for three weeks. Twenty-five B. longicaudatus of mixed life stages were exposed to furfural in 50-ml centrifugation tubes containing solutions of 270, 450, 720, or 990 ppm for 24 hr; or 0, 450, 720, or 990 ppm for 48 hr. The 270 ppm rate was tested for 24 hr only. After exposure, the nematodes, still in their respective suspension, were inoculated into Cone-tainers. The experiment was arranged as a randomized complete block design with six replications. After 30 d, nematodes were extracted from the total soil volume of each Cone-tainer using the centrifugal flotation technique (Jenkins, 1964) and quantified. Population density of B. longicaudatus was subjected to analysis of variance and means were separated according to Fisher's least significant difference (LSD) test ( $P \le 0.05$ ).

Thatch study: This experiment was conducted to quantify effects of thatch on efficacy of furfural. This experiment was a  $2 \times 5$  factorial experiment with two thatch treatments (with or without) and five concentrations of furfural (0, 270,450, 720, and 990 ppm). The experiment was arranged as a randomized complete block design with five replications. To obtain thatchfree turf, 3.8-cm-diam.  $\times$  2.5-cm-deep plugs of recently established 'Tifdwarf' bermudagrass were collected from pots containing no thatch layer or *B. longicaudatus* 



FIG. 2. Effect of thatch on efficacy of furfural against *Belonolaimus longicaudatus* on creeping bentgrass 30 d after treatment averaged across furfural concentrations. Columns represent 50 observations; combined data from two identical trials, each with five concentrations and five replications. Columns with common letters are not different according to Fisher's LSD test ( $P \le 0.05$ ).

using a soil probe. For thatch turf, the same probe was used to remove plugs of 'Tifdwarf' bermudagrass from an established putting green, where no B. longicaudatus were detected, that had a thatch layer  $\geq 2.5$  cm. Twentyfive plugs of each thatch treatment were planted into Cone-tainers, 0.25 cm of sand was added to the surface of each as top-dressing. The turf was allowed to establish a root system for 3 wk before inoculation. Fifty  $\pm 4$  B. longicaudatus were inoculated into each Cone-tainer and allowed to reproduce for 3 wk before treatment. Furfural treatments were applied as a drench in 50 ml of solution. Fifty milliliters is approximately twice the soil pore volume, which is enough to displace the existing soil solution with the furfural solution. After 30 d, nematodes were extracted from the total soil volume in the Conetainer using the modified centrifugal flotation technique and quantified. Nematode data was subject to a two-way analysis of variance with main effect means separated according to Fisher's LSD test ( $P \le 0.05$ ).

*Spray application:* The most common method for liquid turf pesticide applications is surface spray followed by irrigation. Therefore, an experiment was conducted to determine if surface spray application of furfural is an effective method for application of furfural to turf.

Thirty-five Cone-tainers were planted with 'Penncross' creeping bentgrass seeded at 196 kg/ha (0.04 g/ Cone-tainer) and allowed to germinate and establish a root system for 14 d before being inoculated with 50  $\pm$ 4 B. longicaudatus. After 6 wk, furfural solutions (0, 1,350, 2,250, 3,600, and 4,950 ppm) were applied as a spray in 10 ml of solution, the equivalent to a surface spray followed by 0.6 cm of irrigation, a typical method for applying turf pesticides. These rates applied the same amount of furfural per Cone-tainer as used in the thatch experiment, but in five times less solution volume. The experiment was arranged in a randomized complete block design with seven replications. After 30 d, nematodes were extracted from the total soil volume in the Cone-tainer using the modified centrifugal flotation technique and quantified. Nematode data were subject to analysis of variance and treatment means were separated according to Fisher's LSD test ( $P \le 0.05$ ).

## RESULTS

Duration and rate study: All rates of furfural decreased  $(P \le 0.05)$  the number of *B. longicaudatus* recovered compared with the 0 rate (Fig. 1). Treatment effects increased with increasing rates up to 720 ppm of



FIG. 3. Effect of increasing concentration of furfural applied as a soil drench against Belonolaimus longicaudatus on bermudagrass 30 d after treatment averaged across thatch treatment. Columns represent 20 observations; combined data from two identical trials, each with two thatch treatments and five replications. Columns with common letters are not different according to Fisher's LSD test ( $P \le 0.05$ ).

furfural. No B. longicaudatus were recovered following posure times, 24 and 48 hr exposure times were not exposure to  $\geq$  720 ppm furfural. For rates with two ex- different from each other (P > 0.05).



FIG. 4. Effect of increasing concentration of furfural applied as a spray against Belonolaimus longicaudatus on creeping bentgrass 30 d after treatment. Columns represent combined data from two identical trials, each with seven replications. Columns with common letters are not different according to Fisher's LSD test ( $P \le 0.05$ ).

Thatch study: No interaction among main factors was observed (P > 0.1), indicating that thatch did not impair furfural efficacy. Presence of thatch caused a slight decrease ( $P \le 0.05$ ) in population density of *B. longicaudatus* (Fig. 2). Furfural reduced the number of *B. longicaudatus* recovered, and efficacy increased with increasing concentration (Fig. 3).

Spray application: Furfural decreased the number of *B. longicaudatus* recovered. However, nematode population densities from treated Cone-tainers were less than from the 0 ppm rate only for solution concentrations  $\geq$  3,600 ppm (Fig. 4).

#### DISCUSSION

Our results indicate that furfural is an effective nematicide for treatment of *B. longicaudatus* when the nematodes are exposed to sufficient concentrations. Results from the rate and exposure experiment indicate that exposure to 270 ppm of furfural for 24 hr or more can reduce numbers of *B. longicaudatus*, although 450 ppm was required to cause a reduction of 75%. However, achieving these concentrations in turf soil might prove difficult. The current labeled rate for Multiguard Protect EC on turf is 75 liters/ha. Based on a spray application followed by 0.6 cm of irrigation, and assuming even dispersal of the product in the top 7.5 cm of the soil profile, the concentration in soil solution that nematodes would be exposed to would be around 180 ppm.

Thatch, although having a statistical effect, did not appear to affect efficacy to any large degree, but delivery method is likely to affect results. When furfural solution was applied directly to the soil as a drench, nematode reductions were not as great as obtained from direct exposure to solution. The solution volume applied in the thatch experiment was mathematically sufficient to displace existing soil moisture with furfural solution, but it is possible that some dilution was happening. Also, the decomposition rate of furfural in soil, aided by microbial activity and aerobic conditions, is likely to be much higher in the soil than in solution.

Whereas direct comparison between spray and drench treatment was not made, when furfural was applied as a spray nematode reductions were much less than those obtained from drench treatments applying the same amounts of furfural. This is likely, in part, because of dilution that occurs in soil solution. Turf soil always has some amount of water occupying the pore spaces, so the treatments were not applied to dry soil. The existing soil moisture will dilute the treatments, which could affect the results. Also, a spray treatment typically delivers pesticides in a band, so the pesticides are not evenly dispersed in the soil profile. In all likelihood, a higher concentration of furfural was delivered into the upper portions of the Cone-tainers and very little near the bottom.

Although furfural has activity against *B. longicaudatus*, it is impossible to accurately extrapolate greenhouse results to the field. Additional research is required to optimize furfural under field conditions. Future research will evaluate efficacy of spray applications of Multiguard Protect EC to turfgrass infested with *B. longicaudatus* in the field. These future trials will explore varying rates, application intervals, and study effects on nematodes at different soil depths.

### LITERATURE CITED

Busey, P., Giblin-Davis, R. M., and Center, B. J. 1993. Resistance in *Stenotaphrum* to the sting nematode. Crop Science 33:1066–1070.

Cisar, J. L., and Snyder, G. H. 1996. Mobility and persistence of pesticides applied to a USGA green. III: Organophosphate recovery in clippings, thatch, soil, and percolate. Crop Science 36:1433–1438.

Cobb, N. A. 1918. Estimating the nema population of soil with special reference to the sugar beet and root gall nematodes *Heterodera schachtii* Schmidt and *Heterodera radicicola* (Greef) Muller, and with a description of *Tylencholaimus aequalis* n. sp. USDA, Agriculture Techniques Circular 1:47.

Crow, W. T. 2005. How bad are nematode problems on Florida's golf courses? Florida Turf Digest 22:10–12.

Crow, W. T. 2007. Understanding and managing parasitic nematodes on turfgrasses. Pp. 351–374 *in* M. Pessarakli, ed. Handbook of turfgrass management & physiology. Boca Raton, FL: CRC Press.

Giblin-Davis, R. M., Cisar, J. L., Bilz, F. G., and Williams, K. E. 1992. Host status of different bermudagrasses (*Cynodon* spp.) for the sting nematode, *Belonolaimus longicaudatus*. Journal of Nematology 24(suppl.):749–756.

Hensley, J., and Burger, G. 2006. Nematicidal properties of furfural and the development for nematode control in various crops for the United States markets. Journal of Nematology 38:274 (Abstr.).

Jenkins, W. R. 1964. A rapid centrifugal-floatation technique for separating nematodes from soil. Plant Disease Reporter 48:692.

Johnson, A. W. 1970. Pathogenicity and interaction of three nematode species on six bermudagrasses. Journal of Nematology 2:36–41.

Luc, J. E., Crow, W. T., Stimac, J. L., Sartain, J. B., and Giblin-Davis, R. M. 2006. Influence of *Belonolaimus longicaudatus* on nitrate leaching in turf. Journal of Nematology 38:461–465.

Perry, V. G., and Rhoades, H. L. 1982. The genus *Belonolaimus*. Pp.144–149 *in* R. D. Riggs, ed. Nematology in the southern region of the United States. Southern Cooperative Series Bulletin 276. Fayetteville: University of Arkansas Agricultural Publications.

Rodríguez-Kábana, R., Kloepper, J. W., Weaver, C. F., and Robinson, D. G. 1993. Control of plant parasitic nematodes with furfural—A naturally occurring fumigant. Nematropica 23:63–73.

Smiley, R. W., Dernoeden, P. H., and Clark, B. B. 1992. Abiotic agents of noninfectious diseases. Pp. 8–10 *in* Compendium of Turfgrass Diseases, 2nd ed. St. Paul, MN. APS Press.

USGA 1993. USGA recommendations for a method of putting green construction. USGA Green Section Record 31:1–3.

Wierckx, N., Koopman, F., Ruijssenaars, H. J., and de Winde, J. H. 2011. Microbial degradation of furanic compounds: Biochemistry, genetics, and impact. Applied Microbiology and Technology 92:1095– 1105.