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Leaching Behavior of Two Pendimethalin Formulations in a Soilless Growing Medium

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Pendimethalin is commonly applied for PRE weed control in container nursery production. Field and laboratory trials were conducted to determine herbicide effectiveness and leaching of two pendimethalin formulations in pine bark, the primary component used in the growing medium of container-grown plants in the South. The microencapsulated (ME) formulation of pendimethalin gave lower control of southern crabgrass and leached deeper than the emulsifiable concentrate (EC) formulation in pine bark. The EC formulation controlled southern crabgrass 93% compared with only 70% with the ME formulation at 4.48 kg ai ha^{-1}. After applying 3.4 kg ai ha^{-1} pendimethalin and 17.8 cm of irrigation water, the ME formulation showed greater leaching into the 3- to 6- and 6- to 9-cm depths than the EC formulation on the basis of a southern crabgrass bioassay. Using a microwave extraction method, only 0.91 mg kg^{-1} pendimethalin was found in the 3- to 6-cm pine bark depth compared with 4.0 mg kg^{-1} for the ME formulation. Below the 6-cm depth, no pendimethalin was detected when the EC formulation was applied, but 0.5 ppm was found for the ME formulation. No pendimethalin was detected in effluent collected from irrigation water for the EC; however, 3.0 mg kg^{-1} was collected from leachate for the ME formulation. Pendimethalin ME is leaching much deeper than the EC formulation in the pine bark profile, which results in lower weed control. This extensive leaching of ME formulation may be due to capsule movement with the irrigation water, combined with a delayed release of pendimethalin, which then binds to the pine bark much lower in the container profile.

Nomenclature: Pendimethalin; southern crabgrass, *Digitaria ciliaris* (Retz.) Koel.

Key words: Bioassay, dinitroaniline, emulsifiable concentrate formulation (EC), leaching, microencapsulated formulation (ME), soilless media.

A primary means of weed management in container nursery production is PRE herbicide applications. Three to six PRE herbicide applications may be required per year (White and Klingman 2014). Pendimethalin is widely used in the container nursery industry for PRE control of annual grasses and certain broadleaf weeds. Pendimethalin, in the dinitroaniline herbicide class, is a mitotic disrupter that causes root growth inhibition in sensitive species (Shaner 2014). It has a water solubility of 0.275 mg L^{-1} at 25 C and is considered immobile in field soil as it binds to organic matter and clay. The ME formulation of pendimethalin was developed to reduce staining, ease cleanup, reduce odor, and increase effectiveness in no-till crop production (Heinz 2005). These benefits would also be of value to the nursery industry.

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Pendimethalin does not readily leach in field soil. In one study, 90% of the pendimethalin residue in soil was recovered within the upper 10 cm of soil after 45 and 90 d after application of a suspension concentrate formulation of pendimethalin to a loamy sand soil (Lee et al. 2000). In a sandy loam soil, the majority of pendimethalin was recovered from the 0- to 10-cm depth when an EC formulation of pendimethalin was applied (Chopra et al. 2010). However, pendimethalin EC leached to a greater extent in pine bark, a primary component of the media used in container nursery production, than in field soil (Simmons and Derr 2007).

Because of the daily irrigation used in container nursery production, leaching and offsite movement of herbicides is a concern. Excessive leaching of a herbicide could reduce weed control effectiveness, pose an increased risk of root injury to desired crops, and could be an environmental concern if the chemical leaches out of the container.

Formulation may affect resulting weed control with a given active ingredient. Hatzinikolaou et al. (2004) evaluated EC and ME formulations of pendimethalin in a silty clay loam. The EC formulation was more phytotoxic to oat (*Avena*

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sativa L.) than the ME formulation, whereas the ME formulation had slightly longer persistence, perhaps due to a delayed release of the active ingredient. Vasilakoglou and Eleftherohorinos (2003) reported that ME acetochlor caused a higher reduction in oat root growth than EC acetochlor in field soil. Conversely, other studies have indicated no differences in grass control with ME or EC acetochlor in field soil (Doub et al. 1988; Krausz et al. 2000; Mueller and Hayes 1997).

Several studies report movement of pendimethalin EC into the root zone, causing decreased weed control and root injury in soilless mixes (Derr and Salihu 1996; Derr and Simmons 2006; Singh et al. 1981, 1984; Thetford and Gilliam 1991). Application of pendimethalin EC has injured both foliage and roots of certain nursery crops, including azalea (*Rhododendron* spp.), Japanese holly *(Ilex crenata* Thunb.) and ornamental grasses (Derr and Simmons 2006; Skroch et al. 1991; Stamps and Neal 1990).

Limited information is available on the leaching behavior of ME herbicide formulations in pine bark. Factors that influence herbicide effectiveness include rainfall, microbial activity, soil temperature, soil moisture content, porosity, organic matter, clay content, and soil structure and texture (Weber 1990). Differences in physiochemical properties between pine bark and field soil contribute to greater pendimethalin EC movement in pine bark (Derr and Simmons 2006). Pendimethalin formulation may also influence herbicide effectiveness and movement. The objective of this study was to compare the effectiveness of pendimethalin EC and ME in container production using pine bark as the growing medium, to develop a standard curve for southern crabgrass response to the two formulations, to quantify their leaching potential using a southern crabgrass bioassay compared with microwave-assisted extraction and quantification, and to compare weed control effectiveness with their respective leaching patterns.

Materials and Methods

General Methods. Southern crabgrass was used in the trials, as it is a common weed infesting container-grown nursery crops. Southern crabgrass seed was collected from plants grown by the authors. Studies were conducted using 100% loblolly pine (*Pinus taeda* L.) bark, which had a particle size distribution as follows: 1% $<$ 0.053 mm, 31% 0.053 to 1.00 mm, 18% 1.00 to 2.00 mm, 29% 2.00 to 4.76 mm, and 21% $>$ 4.76 mm. For the greenhouse trials, southern crabgrass was seeded at 1.2 ml per pot and grown under a high and low temperature of 28 C and 12 C, respectively. Experimental design was a randomized complete block with four replications for each study. Each experiment was repeated and since there was no treatment-by-trial interaction for any study, data from the two trials were combined into one analysis. Data were subjected to either ANOVA with mean separation using Fisher's LSD ($P = 0.05$) or regression analysis.

Southern Crabgrass Control. Four-liter plastic pots were filled with pine bark and seeded with southern crabgrass at 0.2 ml of seed per pot. The pots were maintained outdoors on a gravel pad during the summer with irrigation applied overhead each day. Two formulations of pendimethalin, EC and ME, were both applied at 2.24 and 4.45 kg ai ha^{-1} after the pots were seeded and compared with nontreated pots. There were three pots per plot. Pots were fertilized with a slow-release product (Osmocote 15–9–12, Everris International B.V., P.O. Box 40, 4190 CA Geldermalsen, the Netherlands) at 10 g per pot. Southern crabgrass control was determined visually on a 0 to 100 scale, with 0 = no control and 100 = complete control, and southern crabgrass plants were counted 25 d after treatment.

Standard Curve. A 30-g sample of pine bark was added to each glass beaker. Pendimethalin EC ad ME were added to obtain concentrations of 0, 0.312, 0.625, 1.25, 2.5, 5, 10, and 20 mg kg^{-1} pine bark. These beakers were shaken thoroughly to mix the applied pendimethalin; pine bark was then placed into 3.8-cm-diam pots. Pots were seeded with southern crabgrass and placed in a greenhouse. After 2 wk, 10 randomly selected plants were removed from pine bark, and root lengths were recorded.

Herbicide Movement as Indicated by Bioassay. Soil columns were constructed of 35-cm increments of polyvinyl chloride pipe, with an internal diameter of 5 cm. Each column was uniformly packed with 7 cm of construction sand, followed by 24 cm of air-dried pine bark. Ends were sealed with a threaded cap and 0.6-cm tubing was used in the center of each cap for drainage. Landscape fabric was placed at the bottom of each end cap. Columns were saturated with water from the bottom up, and allowed to drain for 24 h, at which time the downward movement of water had ceased. Pendimethalin ME and EC at 3.4 kg ai ha^{-1} was then

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surface applied via pipette. After 15 min, each column was irrigated with 20 ml of water. After applying 2.54 cm (51.5 ml) every day for 7 d, the columns were allowed to drain for 24 h. Pine bark was plunged out in the following increments: 0 to 3 cm, 3 to 6 cm, 6 to 9 cm, 9 to 12 cm, 12 to 18 cm and 18 to 24 cm, and placed into 3.8-cm-diam plastic pots. Southern crabgrass was seeded into each pot and grown in a greenhouse. After 2 wk, root weight and root lengths were recorded from five randomly selected plants. Pine bark was removed by rinsing roots in water, with manual removal of large bark particles. Besides ANOVA, data were subjected to Student's *t* test to establish differences in treated and nontreated plants.

Pendimethalin Extraction from Pine Bark. Columns were prepared and treated as per the previous trial. After 17.8 cm of irrigation water was run through each column, pine bark was collected from the following depths: 0 to 3 cm, 3 to 6 cm, and 6 to 24 cm and placed in paper bags. Samples were stored immediately at 4 C, and later transferred to large plastic weighing boats and allowed to air dry at room temperature. Samples were stirred periodically with a metal spatula to ensure uniform drying, and returned to paper bags and kept at 4 C until ground. Each air-dried sample was ground in a Wiley mill (Thomas Scientific, P.O. Box 99, Swedesboro, NJ 08085), allowing pine bark powder to fall directly into a 118-ml Qorpak jar (Qorpak jars with Teflon lined lids #16195-340, VWR, Bridgeport, CT 08014). The mill was disassembled and cleaned; parts were solvent rinsed and oven dried between samples.

Next, all samples received 30 ml of hexane : acetone (1 : 1 by vol), which was added to the microwave vessel. Nonpolar solvents such as hexane do not absorb microwave energy so they must be paired with a polar solvent such as acetone for the sample to heat. The presence of hexane in the extraction solvent system aids in the extraction of nonpolar pesticides like pendimethalin (Armstrong 1999, Shen and Lee 2003). Samples were completely submerged and stirred with a clean spatula to ensure thorough wetting by the solvent. Finally, liners were inserted into sleeves, covered with lids, mounted into support modules, and inserted into the MARS 5 (microwave-accelerated reaction system, CEM Corp., Matthews, NC 28104). The control vessel was equipped with a temperature and pressure probe. Power was set at 300 W (for one to four vessels), 600 W (for five to eight vessels), or 1,200 W (for 9 to14 vessels), with 50% power at 2.1 g cm^{-2} pressure and 115 C for 10 min. After cooling, samples were thoroughly stirred into solvents using clean spatulas and transferred from vessels into a 250-ml flask through 1,573 ½ fast-flow fluted-filter funnels (Schleicher & Schuell 1,573 ½ fast-flow fluted-filter funnels, 10 Optic Ave., Keene, NH 03431) supported by glass powder funnels (#10-346-5C, Fisher Scientific, Pittsburgh, PA 15219). Any remaining sample was scraped into the filter with a spatula. Additional 30 ml of hexane : acetone (1 : 1 by vol) was used to repeatedly rinse vessels, adding rinse to filtrate. Filtrate was evaporated to approximately 1 ml using a rotary evaporator, quantitatively transferred with hexane to 15-ml graduated centrifuge tubes, and evaporated to about 0.5 ml using a nitrogen evaporator (12-position, model 111, Organomation Assoc., Berlin, MA 01503). The extract was brought to a 5-ml final volume with hexane, vortexed, and allowed to equilibrate for at least 1 h. A 1-ml aliquot of each sample was transferred to an autosampler vial for analysis by a gas chromatograph, equipped with a microelectron capture detector (GC/ECD) (Agilent 6890 Plus gas chromatograph, Spectralab Scientific, Inc., 151 Nashdene Rd, Unit 9, Toronto, Ontario M1V 2T3, Canada).

A five-point calibration curve was used, with the highest standard at 5.0 mg kg^{-1} and the lowest standard at 0.01 mg kg^{-1}. Correlation must be 0.99 or above to be acceptable. Method validation consisted of six fortification samples analyzed at the estimated lower limit of method validation consisting of 125 µl of 2.0 µg ml^{-1} pendimethalin in 5 g of ground pine bark. Recoveries ranged from 79 to 120% (standard deviation was 18% or 0.008 mg kg^{-1}) Using a one-tailed *t* test, the limit of detection was 0.03 mg kg^{-1} and the limit of quantitation was 0.08 mg kg^{-1}.

Pendimethalin Extraction from Leachate. This method describes the analysis of pendimethalin in effluent samples collected after applying 3.4 kg ai ha^{-1} pendimethalin and 17.8 cm of water through 61 cm of packed pine bark leaching columns as described above. Immediately after effluent collection, samples were kept in cold storage until laboratory analysis was performed. A Speedisk C_{18}XF (Bakerbond Speedisk C_{18}XF, 50 mm extraction disk #8056-06, J.T. Baker, Phillipsburg, NJ 08865) solid-phase extraction disk was precleaned with ethyl acetate and conditioned with methanol followed by reverse osmosis water using a six-port vacuum manifold

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(Speedisk Manual Expanded Extraction Station, six-port, #8095-06, J.T. Baker) (Cobb et al. 2006). The water sample was passed through the Speedisk under vacuum, after which the disk was air dried for 3 min. Pendimethalin was eluted from the Speedisk with ethyl acetate and the extract dried with sodium sulfate before concentration using a nitrogen evaporator. The sample extract was injected into a GC/ECD. The calibration curve consisted of at least five points, with the highest standard at 0.2 μg ml^{-1} and the lowest standard at 0.001 μg ml^{-1}. Correlation must be 0.99 or above to be acceptable.

This method was tested in triplicate for pendimethalin; the lower limit of method validation was 0.1 μg ml^{-1}. Recoveries ranged from 81 to 87% (standard deviation was 5.15% or 0.004 μg ml^{-1}). Using a one-tailed *t* test, we determined the limit of detection to be 0.003 μg ml^{-1} and the limit of quantitation to be 0.08 μg ml^{-1}.

Results and Discussion

Southern Crabgrass Control. On the basis of stand reduction and control ratings, the EC formulation of pendimethalin gave greater southern crabgrass control at 2.24 kg ha^{-1} than the ME formulation at the same rate (Table 1). A similar response was noted at 4.48 kg ha^{-1}, where the EC formulation controlled southern crabgrass 93%, whereas only 70% control was seen with the ME formulation. The ME formulation is less effective than the EC for weed control in pine bark.

Standard Curve. After treating pine bark with known concentrations of pendimethalin, a standard curve was developed to predict herbicide concentration on the basis of southern crabgrass root length (Figure 1). The following percent reduction in southern crabgrass root lengths were determined

Table 1. Southern crabgrass control and stand 25 d after treatment with two pendimethalin formulations in an outdoor container trial using 100% pine bark as the growing medium.

Pendimethalin formulation	Rate	Percent control[a]	Number per pot
	kg ha^{-1}		
Nontreated		0	92.9
Microencapsulated	2.24	39	42.3
Microencapsulated	4.48	70	12.9
Emulsifiable concentrate	2.24	59	12.8
Emulsifiable concentrate	4.48	93	1.8
LSD		20	15.2

[a] Visual rating based on 0 = no control; 100 = plant death.

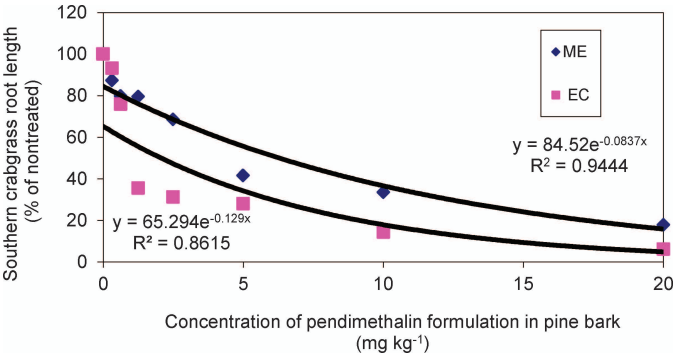


Figure 1. Standard curve for the response of southern crabgrass to progressively higher rates of microencapsulated (ME) and emulsifiable concentrate (EC) formulations of pendimethalin in 100% pine bark growing medium in a greenhouse trial. (Color for this figure is available in the online version of this paper.)

at known pendimethalin concentrations for the ME formulation: 82% (20 mg kg^{-1}), 66% (10 mg kg^{-1}), 58% (5 mg kg^{-1}), 31% (2.5 mg kg^{-1}), 20% (1.25 mg kg^{-1}), 20% (0.625 mg kg^{-1}), and 15% (0.313 mg kg^{-1}). Southern crabgrass root reductions for the EC formulation at known pendimethalin concentrations were: 94% (20 mg kg^{-1}), 86% (10 mg kg^{-1}), 72% (5 mg kg^{-1}), 69% (2. mg kg^{-1}), 64% (1.25 mg kg^{-1}), 24% (0.625 mg kg^{-1}), and 7% (0.313 mg kg^{-1}). The ME formulation displayed lower activity than did the EC formulation of pendimethalin at every concentration. Hatzinikolaou et al. (2004) reported that at 2 mg of pendimethalin kg^{-1} soil, less reduction of oat root length was observed with the ME formulation (67%) than with the EC formulation (83%) at the same concentration.

Pendimethalin Movement. EC pendimethalin leached less than the ME formulation, as it reduced southern crabgrass root weight by 97% in the 0–3-cm depth compared with an 89% reduction in that zone with the ME formulation (Table 2). The ME formulation, however, caused greater reduction in southern crabgrass root weight in the 3- to 6- and 6- to 9-cm depths than the EC formulation. The ME formulation reduced southern crabgrass root weight in the 9- to 12-cm depth, whereas the EC formulation did not reduce root weight when compared with nontreated plants. The ME formulation reduced crabgrass root length by 57% in the 9- to 12-cm depth, whereas the EC formulation caused no reduction at this depth compared with nontreated columns. Hatzinikolaou et al. (2004) reported that the EC formulation of pendimethalin reduced oat root growth greater than the ME

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Table 2. Downward leaching of surface-applied two pendimethalin formulations, microencapsulated (ME) and emulsifiable concentrate (EC), in pine bark columns after irrigating with 2.54 cm of water d^{-1} for 7 d as indicated by a southern crabgrass bioassay in a greenhouse trial.

Depth from surface	Root weight[a]		Root length[b]		Predicted pendimethalin concentration	
	EC[c]	ME[d]	EC[e]	ME[f]	EC[g]	ME[h]
Cm	% of nontreated southern crabgrass				mg kg^{-1}	
0–3	3[i]	11[i]	11[i]	24[i]	13.8	15.0
3–6	24[i]	14[i]	33[i]	29[i]	5.3	12.8
6–9	51[i]	41[i]	76[i]	66[i]	0.0	3.0
9–12	113	57[i]	101	75[i]	0.0	1.4
12–18	100	116	97	101	0.0	0.0
18–24	100	111	99	100	0.0	0.0
LSD	5		2			

[a] Mean untreated root weight 0.23 g.
[b] Mean untreated root length 8.2 cm.
[c] $y = 54.711\ln x - 59.222$ ($r^2 = 0.7596$).
[d] $y = 56.824\ln x - 70.655$ ($r^2 = 0.8266$).
[e] $y = 47.169\ln x - 37.624$ ($r^2 = 0.859$).
[f] $y = 42.305\ln x - 30.33$ ($r^2 = 0.9151$).
[g] $y = 65.29e^{-0.1295x}$ ($r^2 = 0.8615$).
[h] $y = 84.52e^{-0.0837x}$ ($r^2 = 0.9444$).
[i] Significantly less than untreated on the basis of Student's t test (P = 0.05).

formulation in a silty clay loam. EC alachlor and EC acetochlor gave greater weed control than did their ME formulations in field soil (Krausz et al. 2000; Vasilakoglou and Eleftherohorinos 2003).

Initial pendimethalin concentration applied to each column was 27 mg kg^{-1}. Predicted EC pendimethalin concentrations in the 3- to 6-cm depth was 5.3 mg kg^{-1}, whereas at the same depth predicted ME concentration was 12.8 mg kg^{-1} (Table 2). Similar concentrations of ME pendimethalin were found in the 0- to 3-cm and 3- to 6-cm depths. Significantly more pendimethalin is leaching into the 3- to 6-cm depth when the ME formulation was used compared with the EC formulation. Since annual weeds such as southern crabgrass are primarily germinating from the 0- to 3-cm depth, PRE herbicide concentrations need to remain high in that zone. Herbicide leaching below that zone reduced the concentration in the 0- to 3-cm zone, explaining the lower southern crabgrass control seen with the ME formulation compared with the EC formulation (Table 1).

Laboratory Analysis. Pendimethalin concentration recovered from the 0- to 3-cm depth from the pine bark surface was 15.8 mg kg^{-1} for the EC and 12.6 mg kg^{-1} for the ME formulations of pendimethalin (Table 3), consistent with the results from the southern crabgrass bioassay (Table 2). Lab analysis showed higher concentrations of pendimethalin in the 3- to 6-cm depth for the ME than the EC, 4 mg kg^{-1} and 0.91 mg kg^{-1}, respectively (Table 3). Percentage of total pendimethalin recovered at this depth was 5% for the EC and 24% for the ME formulation. No pendimethalin was detected in the 6- to 24-cm depth for the EC formulation; however 0.5 mg kg^{-1} was found in that zone for the ME formulation. Pendimethalin concentration found in water collected from the bottom of the columns (effluent) was 0.003 μg ml^{-1} for the ME, whereas none was detected for the EC formulation. The reduced activity of the ME formulation may be related to the physical properties of the shell material that influences the release rate of herbicide into the soil/water matrix. Average total pendimethalin recovered was 62% for the EC and 67% for the ME formulation. The lower recovery may be due to photodecomposition or microbial breakdown of the herbicide over the study period.

Pendimethalin concentrations were lower in surface layers and higher in deeper depths of pine bark for the ME formulation compared with the EC formulation. Because of the large pore space in pine bark (Simmons and Derr 2007), capsules in the ME formulation may be moving downward via irrigation water, combined with a delayed release of pendimethalin from capsules, resulting in lower weed control and greater movement into the rooting zone. This potentially could cause adverse root effects to sensitive nursery crops. The increased

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Table 3. Pendimethalin concentration at progressively greater depths from the surface after surface application of the microencapsulated (ME) and emulsifiable concentrate (EC) formulations in pine bark columns after applying 3.4 kg ai ha^{-1} pendimethalin and 17.8 cm of irrigation as determined by laboratory analysis.

	Pendimethalin concentration[a,b]		Total pendimethalin recovered[c]	
Depth from surface	EC	ME	EC	ME
cm	mg kg^{-1}		%	
0–3	15.8	12.6	94.8	73.3
3–6	0.91	4.0	5.2	24.0
6–24	ND[d]	0.5	ND	2.7
Effluent	ND	0.003[e]	ND	0.1
LSD	1			

[a] Detection limit in pine bark = 0.03 mg kg^{-1}.
[b] Detection limit in effluent = 0.003 μg ml^{-1}.
[c] Total pendimethalin applied = 6.7×10^{-4} g.
[d] Abbreviation: ND, not determined.
[e] μg ml^{-1}.

leaching of pendimethalin from the ME formulation explains the lower southern crabgrass control seen with that formulation relative to the EC formulation.

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