EFFECT OF SULFURYL FLUORIDE ON THE PINEWOOD NEMATODE IN PINE WOOD

L.D. Dwinell[1]*, E. Thoms[2], and S. Prabhakaran[3]

[1]USDA Forest Service, Athens, GA; [2]Dow AgroSciences LLC, Gainesville, FL; [3]Dow AgroSciences LLC, Indianapolis, IN.

The pinewood nematode (PWN) (*Bursaphelenchus xylophilus*), the causal agent of pine wilt disease, has been intercepted in pine chips, unseasoned pine lumber, and wood packing material (WPM). Likewise, the PWN's insect vectors, *Monochamus* spp. (pine sawyers), have been found in pallets, crates and dunnage. The PWN, which is native to North America, is transmitted to recently killed conifers or freshly felled logs during egg-laying of the insect vector (aka "secondary transmission"). It is through secondary transmission that the PWN gets into the wood transport system.

Several countries now regulate the import of all coniferous chips, logs, sawn wood, and WPM to protect their forest from the PWN and other exotic pests. Various mitigation procedures have been investigated including heat treatments and methyl bromide fumigation. Although methyl bromide can be used for quarantine purposes under the Montreal Protocol, there is interest internationally in finding an alternative fumigant for wood packing material. The potential of sulfuryl fluoride (SF) as an alternative for methyl bromide for managing the PWN and its insect vectors in unseasoned pine has not been evaluated.

This exploratory study was to undertaken to determine if SF could kill the PWN and its insect vectors in unseasoned pine lumber fumigated in chambers.

Materials and Methods. Two laboratory studies were undertaken to determine the accumulated CT (concentration x time) dosage of SF required to eradicate the PWN in shortleaf pine (*Pinus echinata*) sticks. The experiments were conducted at Purdue University in a one-ft^3 fumigation chambers in which chemical concentration and temperature were controlled. In the first experiment, each of the 4 chambers was loaded with 20 2.5 x 2.5 x 25 cm shortleaf pine sticks naturally-infested with the PWN. The temperature was held at 20^oC for the fumigation period of 24 h and the concentration of SF was 30 and 60 g/m^3. Each chamber treatment was replicated twice and twenty sticks served as untreated controls. In the second experiment, SF concentration was held at 60 g/m^3 for 24 h; two temperatures were tested (25^oC and 30^oC). The chamber load was 25 shortleaf pine sticks. Twenty-five sticks served as untreated controls. In both experiments, a thin section of wood was sawn from the center of the sticks and nematodes extracted using the Baermann funnel procedure. The occurrence of the PWN in the sticks was recorded and expressed as percent of sticks positive for PWN. The wood moisture content, expressed on a dry weight basis, was determined by drying a second set of wafers at 105^oC for 24 h.

The efficacy of SF was tested in field fumigation in April 2003 at the Whitehall Forest, Athens, GA. Logs of shortleaf pines that had been killed by the southern pine beetle (*Dendroctonus frontalis*) in 2002 and subsequently colonized by pine sawyers and the PWN were debarked and custom sawn into lumber and cants by Hogan Lumber Company (Cleveland, GA). The logs were salvaged about eight months after the pines had died and sawn into lumber two weeks prior to fumigation. The slabs from the logs, which were sawn into 96.5 cm lengths, were recovered during the milling process. The boards were 2.5 x 15 x 96.5 cm, and 5 x 12.7 x 96.5 cm. The cants were 12.7 x 12.7 x 96.5 cm.

The nine fumigation chambers (volume = 5 m^3) were constructed of lumber and covered with 6 mil polyethylene sheeting. The chambers were on a concrete pad and the edges sealed with wet sand prior to fumigation. Each chamber load consisted of 13 pieces of each of the three lumber sizes and 24 slabs. The introduction of the gas into the chambers was controlled by a digital electronic scale (Ritchie Engineering Corp., Inc) and the SF monitored with a Fumiscope (Key Chemical and Equipment Co.). Because of short half-loss time due to leakage, SF was introduced twice (Table 3). The chamber and ambient air temperature was recorded using Hobo data loggers (Onset Computer Corporation). After fumigation, the boards were sampled for the PWN by drilling two 2.5 cm deep holes with and auger bit and extracting the nematodes from the borings using the Baermann funnel procedure. The slabs, cants and lumber were examined for pine sawyer exit holes in July.

Results and Discussion. In the first laboratory experiment conducted at 20°C, 70% of the shortleaf pine sticks were positive for PWN at 30 SF/m^3 (accumulated CT = 694 g-h/m^3) and 10% were positive at 60g SF/m^3 (accumulated CT = 1393 g-h/m^3,Table 1). However, when SF was held at 60 g/m^3 for 24 h and the temperature increased to 25 and 30°C (accumulated CTs = 1420 and 1426 g-h/m^3, respectively), none of the pine sticks were positive for PWN (Table 2).

In the field experiment, it was necessary to add fumigant after about 4.5 h and the experiment was terminated after about 7.5 h when the target CT dosages were attained. The resulting accumulated CT dosages ranged from 997 to 1751 g-h/m^3 in the fumigation chambers (Table 3). The mean and maximum temperatures for the chambers during fumigation averaged 35.3°C and 40.9°C, respectively. No nematodes were extracted from the lumber in any of the fumigation treatments (Table 4). The results of these laboratory and field experiments indicate that SF can be efficacious in eradicating the PWN in unseasoned lumber destined for the manufacture of wood packing material. Further research, however, is needed to verify these results. Since few pine sawyers exited the non-fumigated slabs this past spring, no conclusions can be made on the efficacy of SF against these insect vectors of the PWN.

Table 1. Results of fumigating PWN-infested shortleaf pine sticks with the PWN in chambers at 30 and 60 g SF/m^3 at 20°C for 24 h.

Treatment[a]	Accumulated CT ($g\text{-}h/m^3$)	% of sticks positive for PWN	
		Before Fumigation	After Fumigation
30 g SF/m3	694	100	70
60 gSF/m3	1393	100	10
Control	0	100	100

[a.] The SF treatments were replicated twice and each replication consisted of 20 sticks. Twenty sticks served as non-fumigated controls. Nematodes were extracted from the samples before and after fumigation using the Baermann procedure.

Table 2. Results of fumigating PWN-infested shortleaf pine sticks in chambers with 60g SF/m^3 at 25 and 30°C for 24 h.

Treatment[a]	Accumulated CT ($g\text{-}h/m^3$)	% of sticks positive for PWN	
		Before Fumigation	After Fumigation
25°C	1420	100	0
30°C	1426	100	0
Control	0	100	100

[a.] The SF treatments were replicated twice and each replication consisted on 25 sticks. Twenty five sticks served as non-fumigated controls. Nematodes were extracted from the samples before and after fumigation using the Baermann procedure.

Table 3. Data on the g/m^3 of SF introduced into the field chambers and the resultant CT dosages ($g\text{-}h/m^3$).

	SF (g/m^3) introduced at time 1 and 2			
Chamber[a]	Time 1[b]	Time 2[b]	Total SF[b]	CT ($g\text{-}h/m^3$)[c]
1	102	357	459	1751
2	102	533	635	1039
3	102	345	447	1538
4	244	78	322	1506
5	153	317	470	1192
6	153	357	510	997

[a.] The chambers had a volume of 5 m^3.
[b.] SF in g/chamber. Time 1 = 0; Time 2 = 4.5 h. Time 2 concentrations increased to compensate for rapid half loss times.
[c.] CT dosages corrected for atmospheric changes (e.g. relative humidity & CO_2) by monitoring non-fumigated controls.

Table 4. Recovery of PWN from unseasoned lumber not fumigated or fumigated with SF.

Lumber Dimensions (cm)[a]	%WMC[b]	% of lumber samples positive for PWN before and after fumigation with SF			
		Non-fumigated		Fumigated	
		Pre-	Post-	Pre-	Post-
2.5 x 15 x 96.5	34	90	89	90	0
5 x 12.7 x 96.5	37	93	83	93	0
12.7 x 12.7 x 96.5	32	84	74	84	0

[a.] Each chamber load consisted of 13 pieces of each dimension. Data is the mean of 6 fumigation chambers and three non-fumigated control chambers.
[b.] Mean wood moisture content (WMC) expressed on a dry weight basis.

This exploratory research was made possible by a grant from Dow AgroSciences LLC.