SULFURYL FLUORIDE FOR QUARANTINE TREATMENT OF PINEWOOD NEMATODE

E. Sousa[1], L. Bonifácio[1], P. Naves[1], M. Lurdes Silva Inácio[1], J. Henriques[1], P. Barbosa[2], M. Mota[2], M. Espada[2], T. Wontner-Smith[3], S. Cardew[3], M. J. Drinkall[4], S. Buckley[5] and E. M. Thoms*[6]

[1]Instituto Nacional dos Recursos Biológicos, I.P.ed. ex- Estação Florestal Nacional Avenida da República, Quinta do Marquês 2780 - 159 Oeiras, Portugal
[2]NemaLab-ICAAM, Dept. Biologia, Universidade de Évora 7002-554 Evora, Portugal
[3]Fumigation and Analytical Methods, The Food and Environment Research Agency Sand Hutton, York, YO41 1LZ, UK
[4] Dow AgroSciences, Latchmore Court, Brand Street, Hitchin, SG5 1NH, UK
[5] Dow AgroSciences, Marco Polo/B-ZAC du Font de l'Orme, 790, Mougins, 06250, France
[6] Dow AgroSciences, 7257 NW 4th Blvd. No 20, Gainesville, Florida, USA

The pinewood nematode [PWN] (*Bursaphelenchus xylophilus* Steiner and Buhrer, 1934, Nickle, 1970) is a serious quarantine pest of forestry trees causing Pine Wilt Disease [PWD]. The pest is widespread in North America and Asia. In 1999 it was first recorded in the European Union (EU) in Portugal (Mota *et al*., 1999). In response to this threat, the European Commission issued Emergency Measures against the dissemination of PWN within or outside the EU including the treatment of wood packaging material in accordance to the International Standard for Phytosanitary Measures (ISPM) No. 15 - Guidelines for Regulating Wood Packaging Material in International Trade.

There are two control measures currently included in the Standard: these are the fumigant methyl bromide (MB) and heat treatment (HT). MB is being phased out of use globally and is no longer registered for use in the EU. Sulfuryl fluoride (SF), a broad-spectrum fumigant for controlling insects and nematodes, is currently under evaluation for inclusion in ISPM No. 15. In the EU, SF has been granted Annex 1 listing under EU Directive 98/8/EC (Biocides) for Product Type 8 (Wood Preservative) and Product Type 18 (Insecticides). Annex 1 listing under EU Directive 91/414 (Pesticides) has been granted with entry into force on the 1st November 2010.

Following efficacy studies by Dwinell *et al*. (2003, 2005) and Flack *et al.* (2008), which showed SF was effective against PWN, a further study was initiated in Portugal in 2010 to validate these results. Maritime pine (*Pinus pinaster*) naturally infested with PWN was collected from the Setúbal Province, Portugal in March 2010 and maintained in incubation chambers at optimum conditions for PWN propagative development (25^{o} C, 75% r.h.). Bioassay wood boards, 15 cm thick x 10 cm wide x 45 cm long, were prepared for SF dosage validation and incubated for 4 days under the same conditions. After this period these conditions were adjusted (12^{o}C, 50% r.h.) for a 7 days to favour the development of the PWN J_{III} (dispersal, resistant juvenile stage). The target of the incubation programme was to increase the density of individuals enable at least 100,000

to be tested per treatment (to meet Probit 9 quarantine standards) with > 60% of the population being present as the resistant stage.

Two 12.2 m long shipping containers were lined with 4 mm thick polystyrene insulation panels (Wallmate^{TM}, The Dow Chemical Company, Midland, MI, USA) and the temperature controlled using thermostatic coil-in-oil electric heaters and air conditioners. A cylinder of commercial grade ProFume$^{®}$ gas fumigant (99.9% sulfuryl fluoride, Dow AgroSciences, Indianapolis, IN, USA) was placed on the electronic weighing scales and connected to the introduction tubes; the required amount of SF was dispensed via a needle valve to achieve target dosages into 1 m^3 polyvinyl chloride fumigation chambers containing the bioassays. Bioassays were fumigated with SF for 24 h at a range of temperatures and dosages: 15^{o}C, 3169 - 4407 g-h/m^3; 20^{o}C, 1901 - 4051 g-h/m^3; 30^{o}C, 1385-2141. Nematodes were extracted using the method described by Penas *et al.* (2002) immediately before and 24h, 72h and 21-42 days after fumigation. Identification and counts of PWN J_{III} and other juveniles and adults were made following extraction under an Olympus BX30 stereoscopic microscope (Olympus Corporation, Tokyo, Japan) and percentage mortality determined. At 21-42 day assessments complete mortality was recorded following fumigations at 15 and 30^{o}C and 94.06-100% at 20^{o}C. The results validate previous research for 15 and 30^{o}C and a requirement for an increase in dosage at 20^{o}C. A proposed treatment schedule is shown Table 1.

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Table I. Proposed treatment schedule for 24 h sulfuryl fluoride fumigation of unseasoned pine for control of pinewood nematode

Mean ºC	Min. Target CT Dosage (g-h/m^3)	SF Dose (g/m^3)	Minimum Concentration (g/m^3) at hour:				
			0.5	2	4	12	24
15-29.9	3200	183	188	176	163	131	93
20-24.9	4400	250	255	240	228	183	130
25-29.9	3200	183	188	176	163	131	93
30 and above	1400	82	87	78	73	58	41