Effects of Some Fumigants on Mortality of the Pine Wood Nematode, *Bursaphelenchus xylophilus*Infesting Wooden Packages

7. Fumigation Schedules for Pine Wood Nematode by Mixture Gas of Methyl Isothiocyanate and Sulfuryl Fluoride

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Abstract: Red pine packing material infested with the pine wood nematode, Bursaphelenchus xylophilus was fumigated at different temperatures and doses of mixture gas of methyl isothiocyanate (MITC) and sulfuryl fluoride (SF) (MITC 30%, SF 30%, carbon dioxide 40%) for 24 hours with 50% loading in a 100-liter fiberglass fumigation box. The data showed that complete mortality was attained at 21 g/ m^3 of MITC and 21 g/m 3 of SF (21 g/m 3 of MITC/SF) at 10 $^{\circ}$ C, 18 g/m 3 of MITC/SF at 15 $^{\circ}$ C and 15 g/m 3 m³ of MITC/SF at 25°C, respectively. A high correlation coefficient (R: 0.996) was observed between CT product and applied dose of SF, and CT products of SF for complete mortality were at each of 482 $\text{mg} \cdot \text{h}/l$ at 10°C , $413 \text{ mg} \cdot \text{h}/l$ at 15°C and $346 \text{ mg} \cdot \text{h}/l$ at 25°C , respectively, while a low correlation coefficient (R: 0.676) was observed between CT product and MITC dose, because of a high sorption ratio to packing material. The data from tarpaulin sheet fumigation showed that complete mortality was attained at each of 27 g/m3 of MITC/SF at 10°C, 27 g/m3 of MITC/SF at 15°C and 15 g/m3 of MITC/ SF at 25°C, respectively. Some survivors, however, were confirmed at 21 g/m³ of MITC/SF at 15°C, because of low gas concentration by gas leakage from the sheet. Doses of MITC/SF in fumigation schedule were determined by adding safety factor (1.17-1.37) to doses calculated with the CT products of SF in the 100-liter fumigation test and the tarpaulin fumigation test. Mixture gas of MITC and SF fumigation schedule for tarpaulin sheet are at each of 33 g/m³ of MITC/SF at 10°C, 27 g/m³ of MITC/SF at 15° C and $21~g/m^3$ of MITC/SF at 25° C for 24 hours with 50% or below loading.

Key words: quarantine treatment, methyl isothiocyanate, sulfuryl fluoride, mixture gas, fumigation, mortality, CT product, *Bursaphelenchus xylophilus*, wood packing material

Introduction

Soma *et al.* (2001) reported that susceptibility of the pine wood nematode, *Bursaphelenchus xylophilus* to sulfuryl fluoride (SF) fumigation showed lower effective against the nematode, and that methyl isothiocyanate (MITC) was absorbed so much to wood packing material. Further, Soma *et al.* (2004) also conducted mortality test on forest insect pests, *Xyleborus perforans* (all stages), *X. pfeili* (all stages), *Callidiellum rufipenne* (adult) and *Cryphalus fulvus* (all stages) by mixture gas of MITC and SF, and complete mortality of all stages of the insects was attained at 15 g/m³ of MITC and 15 g/m³ of SF (15 g/m³ of MITC/SF) for 24 hours at 20°C in tarpaulin sheet fumigation.

Here we report the result of mortality confirmatory tests for the pine wood nematode by mixture gas fumigation of MITC and SF.

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Materials and Methods

Test Wood Packing Material

Red pine, *Pinus densiflora* naturally infested with the pine wood nematode, *Bursaphelenchus xylophilus* (Steiner and Buhrer) Nicklei was collected in Ibaraki and Tochigi Prefectures in April to June 2005. A log was sawed into lumber (10-15 cm thick \times 10-15 cm wide \times 50 cm long). Lumber with more than 10,000 nematodes per 100 g of sample was used for the mortality test. The pine lumber was stored for two to four days at fumigation temperatures until testing.

Fumigant

Mixture gas of MITC and SF (MITC 30%, SF 30% and carbon dioxide 40%) filled in a high-pressurized gas cylinder was used for the test. This type of mixture gas of MITC and SF was registered as agricultural chemical for logs infested with forest insect pests in Japan in 2004.

Fumigation

100-Liter Fumigation Box

The test lumber was placed in a 100-liter fiberglass fumigation box equipped with a gas circulation and ventilation apparatus, ports for gas application and sampling, a manometer and temperature probes, and then fumigation was conducted at different temperatures and doses for 24 hours with 50% loading.

Tarpaulin Sheet

Fumigation was conducted under a tarpaulin sheet in an air-conditioned fumigation chamber. Filler packing material of red pine lumber ($15~\text{cm}\times15~\text{cm}\times100~\text{cm}$, 18~lumber; and $10~\text{cm}\times10~\text{cm}\times100~\text{cm}$, 9~lumber) and red pine board ($3~\text{cm}\times15~\text{cm}\times100~\text{cm}$, 90~boards); and $2~\text{cm}\times10~\text{cm}\times100~\text{cm}$, 90~boards) were loaded in the size of $1~\text{m}^3$ ($1~\text{m}\times1~\text{m}\times1~\text{m}$). The test material infested with the pine wood nematode was also placed in the center of the load. A plastic pipe frame ($1.25~\text{m}\times1.25~\text{m}\times1.25~\text{m}$) was placed over the load, and then the frame was covered with an EVO sheet (Bariastar) of 0.1~mm thickness. Sand snakes were placed on the sheet skirt to prevent gas leakage from the sheet. The capacity of the tarpaulin sheet was $1.95~\text{m}^3$, and the load factor was 51.2%~(v/v) in the test.

A gas dosing Teflon pipe and temperature sensors were fixed to top and bottom in air space and between lumber of the load. A small-size air circulation fan was placed under the tarpaulin sheet. Mixture gas of MITC and SF was introduced to the tarpaulin sheet by connecting a stainless steel pipe fixed to the top layer of the load. A gas circulation fan was operated for 30 minutes during dosing. Air-fumigant mixture was exhausted for one hour after fumigation. Fumigated material was placed in netted bags and was stored at ambient temperature until the evaluation of mortality.

Measurement of Temperature, Gas Concentration and Moisture Content

Temperatures were monitored with an automatic temperature recorder (Hybrid recorder, AH, Chino). Gas concentrations were monitored at time intervals of 1, 2, 4 and 24 hours during fumigation with gas chromatography (Shimazu) with detector of FID for MITC and TCD for SF. The residual gas ratio was calculated with the following formula;

Residual gas ratio (%)=
$$\frac{100 \times \text{gas concentration after 24 hour fumigation (mg/l)}}{\text{applied dose (g/m}^3)}$$

CT product was calculated with the following formula;

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CT product (mg \cdot h/l) = 1.5C_1 + 1.5C_2 + 11C_4 + 10C_{24} (where C_x is the gas concentration after x hour fumigation).
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Moisture content was measured by the weight difference of samples of lumber and board

pieces before and after drying at 115° C for 18-24 hours. The moisture content of filler lumber and board was $13.6\pm3.4\%$.

Evaluation of Mortality

Before and at 6–7 days after fumigation, the number of nematodes was confirmed by the Bermann funnel method using wood samples fumigated and non-fumigated wood material. Small pieces from a few places of lumber and board were cut off at a still smaller size (3 mm \times 3 mm \times 5 mm) with a specially designed cutting instrument and scissors. A 20–40 g of sample was placed in a funnel, and then the funnel was stored for 48 hours at room temperature. The number of surviving nematodes was counted under a microscope.

Results and Discussion

100-Liter Fumigation Box Gas Concentration

Figure 1 shows average progressive gas concentrations for red pine packing material fumigated at 27 g/m^3 of MITC/SF at 15°C with 50% loading. A high sorption ratio of MITC was confirmed at just after dosing at each temperature, and gas concentrations reduced to as low as 1 mg/l in 4 hours and 0.5 mg/l after 24 hours, while a low sorption ratio was observed on SF fumigation with 25 mg/l after 24 hours.

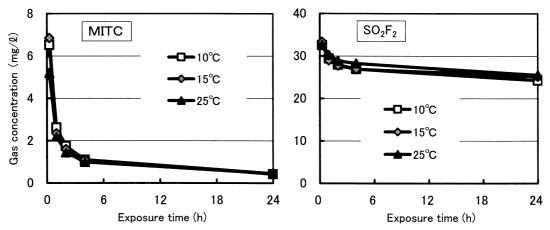


Fig. 1. Progressive gas concentrations for red pine wood packing materials fumigated with mixture gas of methyl isothiocyanate and sulfuryl fluoride for 24 hours with 50% loading.

Mortality and CT Product for Complete Mortality

Table 1 shows mortality of the pine wood nematode fumigated at different temperatures and doses for 24 hours with 50% loading. Table 2 also shows gas concentrations at the end of fumigation, the residual gas ratio and CT products.

Complete mortality was attained at each of 21, 27 and 33 g/m³ of MITC/SF at 10° C, 18, 21 and 27 g/m³ of MITC/SF at 15° C and 15 g/m³ of MITC/SF at 25° C, while some survivors were confirmed at 9 g/m³ of MITC/SF at 25° C in one of three replicate tests.

In SF fumigant, a high residual gas ratio of 87.0-98.7% and an extremely low sorption ratio were confirmed. CT products for complete mortality were at each of $482 \text{ mg} \cdot \text{h}/l$ at 21 g/l of MITC/SF at 10°C , $413 \text{ mg} \cdot \text{h}/l$ at 18 g/m^3 of MITC/SF at 15°C and $346 \text{ mg} \cdot \text{h}/l$ at 15 g/m^3 of MITC/SF at 25°C . A high correlation coefficient (R: 0.996) was observed between the CT product and the applied dose, which the CT products of SF could possibly be used as an indicator of mortality efficacy. In MITC fumigant, residual gas ratios of 1.5-3.9% and an extremely high sorption ratio were confirmed. A low correlation coefficient (R: 0.676) was observed between CT product and applied dose, because of a high sorption ratio, which the CT product of MITC could not be used as an indicator of mortality efficacy.

Table 1. Mortality data for the pine wood nematode infesting red pine packing materials fumigated with mixture gas of methyl isothiocyanate and sulfuryl fluoride for 24 hours with 50% (v/v) loading in a 100 l fiberglass box.

Т			NI f	Bef	ore fumiga	ntion	Af	ter fumiga	tion	
Temper- ature (°C)	Dose ¹⁾ (g/m ³)	I	No. of test lumber	Moisture content ²⁾ (%)	Weight of board ³⁾ (g)	No. of nematode ⁴⁾ per 100 g	Moisture content ²⁾ (%)	Weight of board ³⁾ (g)	No. of nematode ⁴⁾ per 100 g	Survivor (%)
	21	2	4	22.4	124.0	18,300	19.1	820.0	0	0
	27	1	2	24.5	60.9	22,700	18.2	197.4	0	0
10	33	1	2	24.5	60.9	22,700	20.0	198.1	0	0
	Cont.	_	5	23.8	132.9	20,600	19.3	123.9	36,900	179.1
	18	2	4	38.2	191.5	19,800	20.5	811.9	0	0
	21	1	2	38.4	148.5	28,800	27.8	411.0	0	0
15	27	1	2	37.2	40.6	18,900	14.0	411.2	0	0
	Cont.	_	6	36.0	213.0	21,800	15.3	145.7	33,400	153.2
25	9	3	6	28.2	184.7	28,000	17.6	1,015.9	3.45	0.012
	15	3	8	31.8	176.8	28,700	24.3	1,011.1	0	0
-	Cont.	_	7	31.7	269.2	30,600	19.3	165.0	41,000	134.0

¹⁾ Same dose of methyl isothiocyanate and sulfuryl fluoride.

Table 2. Residual gas concentration, residual gas ratio and CT product for the red pine packing material infested with the pine wood nematode fumigated with mixture gas of methyl isothiocyanate and sulfuryl fluoride for 24 hours with 50% (v/v) loading in a 100 l fiberglass box.

			Methy	l isothiocyan	ate	Sulfuryl fluoride					
Temper- ature	Dose ¹⁾	Repli-	Residual gas	Ratio of CT		Residual gas	Ratio of	CT product ³⁾			
(°C)	(g/m^3)	cate	concentration	residual gas ²	product ³⁾	concentration	$residual\ gas^{2)}$	Average	Minimum		
, ,			(mg/l)	(%)	$(\text{mg} \cdot \text{h}/l)$	(mg/l)	(%)	$(\text{mg} \cdot \text{h}/l)$	$(\text{mg}\cdot \mathbf{h}/l)$		
	21	2	0.4	1.9	22.2	19.7	93.8	492	482		
10	27	1	0.4	1.5	23.0	23.5	87.0	651	_		
	33	1	0.7	2.0	35.4	31.5	95.5	820	_		
	18	2	0.4	2.0	20.5	16.8	93.3	426	413		
15	21	1	0.6	2.8	30.7	18.7	89.0	481	_		
	27	1	0.6	2.3	32.1	26.1	96.7	659	_		
25	9	3	0.3	3.3	14.0	8.2	90.6	214	205		
۷۵	15	3	0.6	3.9	31.0	14.8	98.7	285	346		

¹⁾ Same dose of methyl isothiocyanate and sulfuryl fluoride.

Tarpaulin Sheet Fumigation

Gas Concentration, Residual Gas Ratio, and CT Product

Figure 2 shows the progressive gas concentration at $21~\rm g/m^3$ of MITC/SF at $15^{\circ}{\rm C}$ for $24~\rm hours$ with 51.2% loading. The MITC concentration between lumber of the load was low at the beginning of fumigation, and the difference between air space and the load was 57% at 4 hours and 63% at 24 hours. The difference did not correspond to the end of fumigation at 24 hours. The residual gas ratio after 24 hours was as low as 2%, because of high sorption ratio to packing material, while the residual gas ratio of SF was 96% and 64.8% at 4 hours and at 24 hours, respectively, with no difference of gas concentration between air space and lumber of the load. The data indicate that SF is highly penetrating and a low sportive fumigant to packing material.

²⁾ Average moisture content in test board.

³⁾ Weight of wood samples used to detect nematode.

⁴⁾ Number of survivor of the pine wood nematode.

 $^{^{2)}}$ 100×gas concentration after 24 hours fumigation (mg/l)/applied dose (g/m 3).

³⁾ $1.5C_1 + 1.5C_2 + 11C_4 + 10C_{24}$ (C_x : gas concentration after x hours).

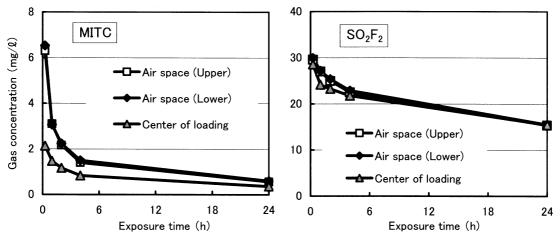


Fig. 2. Progressive gas concentrations for red pine packing materials fumigated with mixture gas at 21 g/m³ of methyl isothiocyanate and at 21 g/m³ of sulfuryl fluoride at 15°C with 51.2% loading.

The residual gas ratio and CT product and mortality of the pine wood nematode are shown in Tables 3 and 4, respectively. The residual gas ratio of MITC was 1.2-3.3%, because of a high sorption ratio to packing material. The CT product was also low values at $14.8-39.2~{\rm mg}\cdot {\rm h}/{\it l}$, which values were not much different compared with the value from the test in a 100-liter fumigation box. The data indicate slight gas leakage of MITC from the tarpaulin sheet, while the residual gas ratio of SF was 35.2-85.3% in tarpaulin fumigation, and the ratio was lower than that of 87% or more in the 100-liter fumigation box (Table 2). This indicates a low sorption ratio to packing material and easy gas leakage from the tarpaulin sheet. Residual gas ratios were lower than those at low temperatures, probably because gas leakage from the sheet was accelerated by wind from air-conditioning of the fumigation chamber, as the air-conditioning would be operated for longer time at a low temperature. Complete mortality was attained at each of 27, 33 and 42 g/m³ of MITC/SF at 10° C, $27~{\rm g/m}^3$ of MITC/SF at 15° C and $15, 21~{\rm g/m}^3$ of MITC/SF at 25° C, while

Table 3. Residual gas concentration, residual gas ratio and CT product for the red pine packing material infested with the pine wood nematode fumigated with mixture gas of methyl isothiocyanate and sulfuryl fluoride for 24 hours with 51.2% (v/v) loading under a tarpaulin sheet¹.

			Methy	l isothiocyana	te		Sulfuryl fluoride			
Temper- ature	Dose ²⁾	1	Residual gas	Ratio of	СТ	Residual gas	Ratio of	CT product ⁴⁾		
(°C)	(g/m ³)	cate	concentration (mg/l)	residual gas ³⁾ (%)	$product^{4)}$ $(mg \cdot h/l)$	concentration (mg/l)	residual gas ²⁾ (%)	Average $(mg \cdot h/l)$	$\begin{array}{c} \text{Minimum} \\ (\text{mg} \cdot \text{h}/l) \end{array}$	
	27	1	0.4	1.5	20.6	9.5	35.2	444	_	
10	33	1	0.6	1.8	30.6	16.0	48.5	594	_	
	42	2	0.8	1.8	39.2	17.1	40.7	702	689	
	15	$2^{5)}$	0.5	3.3	24.7	9.2	61.3	286	266	
15	21	1 ⁵⁾	0.4	1.9	20.9	13.5	64.3	398	_	
10	21	3	0.3	1.4	14.8	16.1	76.7	488	436	
	27	2	0.7	2.6	37.5	18.3	67.6	531	513	
0.5	15	2	0.4	2.7	19.8	12.8	85.3	342	292	
25	21	2	0.3	1.2	15.6	15.6	74.3	478	415	

 $^{^{1)}}$ Test and filler lumber and board were loarded under a tarpaulin sheet of 1.95 m 3 .

²⁾ Same dose of methyl isothiocyanate and sulfuryl fluoride.

³⁾ 100×gas concentration after 24 hours fumigation (mg/l)/applied dose (g/m³).

⁴⁾ $1.5C_1 + 1.5C_2 + 11C_4 + 10C_{24}$ (C_x : gas concentration after x hours).

⁵⁾ Test lot confirmed surviving pine wood nematode.

Table 4. Mortality data for the pine wood nematode infesting red pine packing material fumigated with mixture gas of methyl isothiocyanate and sulfuryl fluoride for 24 hours with 51.2% (v/v) loading under a tarpaulin sheet¹⁾.

T			No. of	Ве	fore fumiga	ition	After fumigation				
Temper- ature (°C)	$\frac{Dose^{2)}}{(g/m^3)}$	Repli- cate	test lumber	Moisture content ³⁾ (%)	Weight of board ⁴⁾ (g)	No. of nematode ⁵⁾ per 100 g	Moisture content ³⁾ (%)	Weight of board ⁴⁾ (g)	No. of nematode ⁵⁾ per 100 g	Survivor (%)	
	27	1	4	17.6	120.4	97,400	21.9	707	0	0	
10 -	33	1	3	23.5	75.0	32,100	19.2	624	0	0	
	42	2	7	20.0	195.6	63,100	20.0	1,309	0	0	
	Cont.		9	20.4	255.7	48,900	14.6	246	46,700	95.5	
	15	2	6	25.0	185.0	18,600	15.4	1,233	77.04	0.413	
15	21	1 3	3 9	23.9 25.7	93.5 212.1	11,500 59,500	16.3 19.6	631 1,724	0.45 0	0.004 0	
	27	2	8	38.0	314.9	22,700	32.7	1,624	0	0	
-	Cont.		13	26.0	433.7	62,300	13.7	265	47,600	76.4	
	15	2	6	23.2	142.9	22,700	20.5	1,231	0	0	
25	21	2	8	16.1	240.8	22,600	19.7	1,444	0	0	
_	Cont.		10	19.8	270.4	22,500	13.4	255	26,300	116.9	

¹⁾ Test and filler lumber and board were loarded in a tarpaulin sheet of 1.95 m³.

some survivors were confirmed at 15 g/m^3 in two replicates and at 21 g/m^3 in one of four replicate tests at 15°C , because the CT product of SF was lower than that $(413 \text{ mg} \cdot \text{h/l})$ in the 100-liter fumigation box (Tables 1 and 2).

Some survivors (3.1–29.0 nematodes per 100 g of wood sample) of free-living nematode (*Rhabditidae Gen. et* sp. and *Plectidae Gen. et* sp.) were observed on all the fumigation schedule. However, these surviving free-living nematodes could be disregarded, as they are non-quarantine pests.

Mortality and Gas Leakage from the Tarpaulin Sheet

Progressive gas concentration, residual gas ratio and CT product of SF and mortality of the pine wood nematode fumigated at 15°C for 24 hours with 51.2% loading are shown in Fig. 3 and Table 5, respectively.

The gas concentration at 27 g/m^3 of SF was high just after dosing, and then the concentration decreased rapidly because of gas leakage, while a high gas concentration was confirmed at 15 g/m^3 of SF, because of low gas leakage. Residual gas ratios were 15.9% at 27 g/m^3 and 94.7% at 15 g/m^3 . However, the CT product of SF at 27 g/m^3 was fairly low when based on the gas reduction curve in Fig. 2, although a little difference of CT was confirmed between $394 \text{ mg} \cdot \text{h/l}$ at 27 g/m^3 and $393 \text{ mg} \cdot \text{h/l}$ at 15 g/m^3 .

Some survivors of the nematode were found at 27 g/m^3 of MITC/SF, and complete mortality was attained at 15 g/m^3 of MITC/SF. These data indicate that complete mortality would not be attained even at a high dose of SF, if gas leakage from the tarpaulin sheet is not prevented during fumigation.

²⁾ Same dose of methyl isothiocyanate and sulfuryl fluoride.

³⁾ Average moisture content in test board.

⁴⁾ Weight of wood samples used to detect nematode.

⁵⁾ Number of survivor of the pine wood nematode.

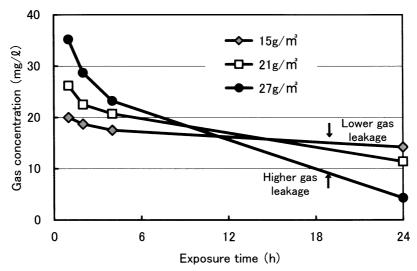


Fig. 3. Relation between sulfuryl fluoride concentration and gas leakage during fumigation of red pine packing materials at 15°C with 51.2% loading.

Table 5. Residual gas ratio, CT product and mortality for the pine wood nematode infesting red pine packing material fumigated with mixture gas of methyl isothiocyanate and sulfuryl fluoride for 24 hours at 15° C with 51.2% (v/v) loading under a tarpaulin sheet¹⁾.

	Methyl isoth	niocyanate	Sulfuryl	fluoride	Mortality			
Dose ²⁾ (m ³)	Ratio of residual gas ³⁾ (%)	CT product ⁴⁾ (mg·h/l)	Ratio of residual gas ³⁾ (%)	CT product ⁴⁾ (mg·h/l)	No. of test wood	No. of nematode ⁴⁾ per 100 g	Survivor (%)	
15	2.7	19.8	94.7	393	3	0	0	
21	1.9	16.9	52.9	436	3	0	0	
27	2.6	35.1	15.9	394	4	514	2.04	

 $^{^{1)}}$ Test and filler lumber and board were laorded in a tarpaulin sheet of 1.95 m 3 .

Proposed Fumigation Schedule and Fumigation Method

Table 6 shows a proposed fumigation schedule of mixture gas of MITC and SF (temperature, dose, exposure time, load factor, minimum gas concentration and minimum CT product). Minimum CT products of SF for complete mortality of the pine wood nematode were $482 \text{ mg} \cdot \text{h}/l$ at 10°C , $413 \text{ mg} \cdot \text{h}/l$ at 15°C and $346 \text{ mg} \cdot \text{h}/l$ at 25°C , respectively based on the value from the test in the 100-liter fumigation box (Tables 1 and 2). Applied doses were at 33 g/m^3 of MITC/SF at 10°C , 27 g/m^3 of MITC/SF at 15°C and 21 g/m^3 of MITC/SF at 25°C , which these doses of MITC/SF in fumigation schedule were determined by adding safety factor (1.17–1.37) to doses at 28.2 of

Table 6. Proposed mixture gas of methyl isothiocyanate and sulfuryl fluoride fumigation schedule for wood packing material infected with the pine wood nematode.

Temperature (°C)	Exposure	Load factor - (%)	Dose		Minimum	Minimum CT		
	time (h)		$MITC$ (g/m^3)	$\frac{SF}{(g/m^3)}$	1 h (mg/ <i>l</i>)	4 h (mg/ <i>l</i>)	24 h (mg/ <i>l</i>)	product of SF $(mg \cdot h/l)$
10-14.9			33	33	29	25	12	490
15-24.9	24	50 or below	27	27	24	21	11	420
25 or above			21	21	19	17	10	350

[•] An air circulation fan is operated for more than 30 minutes during dosing.

²⁾ Same dose of methyl isothiocyanate and sulfuryl fluoride.

 $^{^{3)}}$ $100 \times gas$ concentration after 24 hours fumigation (mg/l)/applied dose (g/m 3) .

⁴⁾ $1.5C_1 + 1.5C_2 + 11C_4 + 10C_{24}$ (C_x : gas concentration after x hours).

MITC/SF at 10°C, 20.4 of MITC/SF at 15°C and 15.3 of MITC/SF at 25°C in the 100-liter fumigation test and the tarpaulin fumigation test.

Minimum gas concentrations at 1, 4 and 24 hours and the CT product were determined with gas consecration records in tarpaulin sheet fumigation.

To obtain enough CT products for complete mortality of the pine wood nematode, a circulation fan should be operated for 30 minutes at start of fumigation and be taken care of gas leakage from the tarpaulin sheet.

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和 文 摘 要

マツノザイセンチュウ Bursaphelenchus xylophilus が寄生した 梱包材のくん蒸剤による消毒試験

7. メチルイソチオシアネート・フッ化スルフリル混合ガス を用いたくん蒸試験

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メチルイソチオシアネート (MITC) とフッ化スルフリ ル (SF) の混合ガスくん蒸剤 (MITC 30%、SF 30%、 炭酸ガス 40%) を用い、マツノザイセンチュウが寄生 した赤松梱包材を 100 l のくん蒸箱で収容率 50%、24 時間くん蒸し、殺虫効果と CT 値の関係を調査した。そ の結果、10℃ では MITC 21+SF 21 g/m³、15℃ で は MITC 18+SF 18 g/m³ 及び 25°C では MITC 15+ SF 15 g/m³ で完全殺虫が得られた。SF 薬量と CT 値 の間には高い相関 (R: 0.996) が認められ、完全殺虫が 得られたときの SF の CT 値は 10℃ で 482 mg·h/l、 15°C で 413 mg·h/l 及び 25°C では 346 mg·h/l であった。MITCは梱包材へのガス収着性が大きく、 CT 値と薬量との相関は低かった (R: 0.676)。 マツノザ イセンチュウが寄生した赤松梱包材 (10~15 cm×10~ 15 cm×50 cm) を天幕くん蒸 (内容積 1.95 m³、収容 率 51.2%、24 時間、投薬から30 分間かくはん) した

結果、 10° 及び 15° では MITC 27+SF 27 g/m^3 、 25° では MITC 15+SF 15 g/m^3 で完全に殺虫された。しかし、 15° では MITC 21+SF 21 g/m^3 で完全 殺虫が得られなかった。これは、天幕からガスが漏洩し濃度が低下したことによるものであった。

天幕くん蒸における温度別薬量基準は、100~l のくん蒸箱を用いた試験で完全殺虫されたときの SF の CT 値を基準に、天幕くん蒸の CT 値から完全殺虫に必要な薬量を算出し、安全係数 $(1.17\sim1.37)$ を乗じた値から設定した。 10° C では MITC 33+SF $33~g/m^3$ (混合ガス $110~g/m^3$)、 15° C では MITC 27+SF $27~g/m^3$ (混合ガス $90~g/m^3$)及び 25° C では MITC 21+SF $21~g/m^3$ (混合ガス $70~g/m^3$)が必要な薬量であり、くん蒸時間は 24 時間、収容率は 50% 以下で、投薬後は天幕内を 30 分間以上かくはんすることが必要である。