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

4. Mortality and CT Product in Methyl Bromide Fumigation with High Loading of Wood Packing Materials

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Abstract: Red pine lumber (15 cm \times 15 cm \times 40 cm) with moisture contents of 17–20% was fumigated with methyl bromide at 50, 60, 70 and 80 g/m³ for 24 hours at 15°C with 54% loading in a 100 liter fiber-glass fumigation box. The result showed that average CT products (mg·h/l) were 455 at 50 g/m³, 533 at 60 g/m³, 604 at 70 g/m³ and 656 at 80 g/m³, respectively. A complete mortality of the pine wood nematode was attained at 80 g/m³ with CT product of 656 mg·h/l and a high dose of methyl bromide would be necessary for a complete mortality of pine wood nematode, *Bursaphelenchus xylophilus* in fumigation with high loading of wood packing materials with low moisture contents. The data from tarpaulin fumigation showed that the nematode would be killed completely if no gas-leakage was observed on tarpaulin fumigation.

Key words: quarantine treatment, methyl bromide, *Bursaphelenchus xylophilus*, mortality, gas sorption, CT product, wood packing material

Introduction

Soma $et\,al.$ (2003) reported that complete moralities of pine wood nematode, $Bursaphelenchus\,xylophilus$ (Steiner and Buhere) Nickle infesting red pine, $Pinus\,densiflora$ were attained for 24 hours at doses of $112\,\mathrm{g/m^3}$ of methyl bromide (MB) at $10^{-1}4.9^{\circ}\mathrm{C}$ (CT product: $900\,\mathrm{mg}\cdot\mathrm{h/l}$), $80\,\mathrm{g/m^3}$ at $15^{-1}9.9^{\circ}\mathrm{C}$ (CT product: $700\,\mathrm{mg}\cdot\mathrm{h/l}$), $64\,\mathrm{g/m^3}$ at $20^{-2}4.9^{\circ}\mathrm{C}$ (CT product: $600\,\mathrm{mg}\cdot\mathrm{h/l}$) and $48\,\mathrm{g/m^3}$ at $25^{\circ}\mathrm{C}$ or above (CT product: $500\,\mathrm{mg}\cdot\mathrm{h/l}$), respectively. They also indicated that shortage of minimum CT products may be sometimes observed on tarpaulin fumigation because of much MB sorption by high loading of wood packing materials with lower moisture content in addition to gas leaky facility, and that additional dose or extension of fumigation time should be taken for attaining minimum CT products.

Here we report the result of further tests for gas sorption, CT product and mortality of the nematode with high loading of Japanese red pines and New Zealand radiator pines.

Materials and Methods

1. Test wood packing material

Red pine logs, *Pinus densiflora* naturally infested with the pine wood nematode were

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collected in Ibaraki Prefecture in April 2003 and sawn into lumber ($15 \text{ cm} \times 15 \text{ cm} \times 40 \text{ cm}$) for mortality tests and New Zealand radiator pine lumber ($15 \text{ cm} \times 15 \text{ cm} \times 100 \text{ cm}$) and board ($1.2-4 \text{ cm} \times 9-14 \text{ cm} \times 90 \text{ cm}$) and Japanese red pine board ($3-15 \text{ cm} \times 15 \text{ cm} \times 100 \text{ cm}$) with different moisture contents were used for gas sorption tests.

2. Fumigation

Mortality test

The lumber and board were stored at fumigation temperatures for 1 to 3 days, and then put in a 100 liter fiber-glass fumigation box $(40 \text{ cm} \times 50 \text{ cm} \times 50 \text{ cm})$ equipped with a circulation fan, ventilation apparatus, and ports for gas application and sampling, a manometer and temperature probes. MB (purity; 99% or more) was introduced with a gas-tight syringe and then fumigated at 50, 60, 70 and 80 g/m³ for 24 hours at 15°C with 54% loading. Air-fumigant mixture was exhausted for one hour after fumigation. Fumigated wood packing materials were placed in netted bags and stored at ambient temperature until detection of the nematode by Bermann funnel method.

Gas sorption test

The plastic pipe frame was placed over the test wood bundle and covered the frame with polyethylene sheet of 0.015 mm in thickness. Pressing of the tarpaulin hems were done by laying sand bags to protect gas leakage. A gas dosing Teflon pipe was fixed to the top layer of the bundle. A temperature probe and a gas sampling Teflon pipe were also fixed between lumbers and boards of the bundle. MB was collected in a graduated dispenser from a cylinder and then introduced into the tarpaulin by connecting dosing pipe fixed to the top layer of the bundle. Fumigation was conducted with different load factors, doses and temperatures.

3. Measurements of gas concentration, temperature and moisture content

Gas concentrations during fumigation were monitored periodically with automatic infrared gas measurement device and interference refractometer and temperatures were also monitored with an automatic temperature recorder. Small size of wooden pieces (0.5 m \times 0.5 mm \times 0.8 mm) of 3 lumbers and boards were used for measurement of moisture content which were determined by the difference of the weight of the sample before and after drying at 120°C for 24 hours.

4. Evaluation of mortality

The number of the nematode in the test wood was confirmed before fumigation and in 6–7 days after fumigation. Pieces of wood from a few places of fumigated or untreated lumbers and boards were collected by sawing and then cut into the size of $3 \text{ mm} \times 4 \text{ mm} \times 5 \text{ mm}$ with scissors. The sample of 30g per place was detected by Bermann funnel method for 24 hours at room temperature for 48–72 hours and then the number of survival nematode was confirmed under microscopes.

Results and Discussion

Mortality test

Table 1 shows progressive gas concentrations and CT products for the lumber with moisture contents of 17–20% fumigated at 4 doses of MB for 24 hours at 15°C with 54% loading. Gas concentrations in each dose were reduced rapidly in 1–6 hour after dosing and were extremely low at the end of fumigation. The CT product was also low because of low gas concentrations at the end of fumigation. Such fumigation conditions of high

loading, low moisture contents and use of short length of lumber (40 cm) may cause lower gas concentrations and CT products.

Table 2 also showed that in tarpaulin fumigation, a complete mortality was also confirmed at 80 g/m³ with CT product of 634-679 mg·h/l, while survived nematode were confirmed on the test lot with CT product of 610 mg·h/l. Soma et al. (2001) reported that minimum CT product for a complete mortality of the pine wood nematode was 650 mg·h/ l at 50 g/m^3 for 24 hours at 15°C with 25°M loading, while in this tarpaulin fumigation, average CT product of 455 mg·h/l was attained at 50 g/m³ for 24 hours at 15°C with 54% loading and moisture contents of 17-20% of the test wood. The data indicated that lower loading of wood packing materials should be applied for practical fumigation for attaining minimum CT product for a complete mortality.

Gas sorption test in tarpaulin fumigation

Figure 1 shows progressive gas concentration for New Zealand radiator pine board fumigated at 48 g/m³ for 24 hours at 26.5-33.5°C with 50 and 70% loading with circulation fan at the beginning of fumigation. Gas reduction curves in 4 replicated tests were almost the same pattern and CT products in 4 replicate tests were 620-680 mg \cdot h/l, which were fulfilled minimum CT product of 500 mg·h/l for a complete mortality of the pine wood nematode at 25°C or above in the test conducted by Soma et al. (2003). Figure 2 also shows progressive gas concentration for Japanese red pine lumber fumigated at 80 g/m³ for 24 hours at 15°C with 25 and 50% loading without circulation fan. Gas reduction curves in 4 replicated tests were also almost the same pattern except for replicate 1 which was lower concentration in first one hour after dosing. The uniformity of gas concentrations was delayed in 0.5 to 4 hours after dosing in fumigation without circulation fan when comparing gas reduction curves in fumigation with circulation fan as shown in Fig. 2.

Table 1.	Progressive	gas	concentrations	and	СТ	products	in	methyl	bromide	fumigation1)	of	red	pine
	lumber ²⁾ .												

Dose (g/m³)	Repli-		Ga	Moisture	CT product				
	cate	1	2	4	6	16	24	content (%) ³⁾	${\sf mg}\!\cdot\!{\sf h}/l$
	1	33.6	26.2	21.2	19.4	16.2	15.2	17.4	456
50	2	32.4	26.1	21.8	19.5	15.7	14.6	19.6	448
	3	28.7	26.2	21.6	_	-	14.1	17.5	461
	Average	31.6	26.2	21.5	19.5	16.0	14.3	18.2	455
	1	39.5	32.6	26.7	24.5	19.8	19.0	20.0	563
60	2	30.8	28.4	22.7	_	_	16.5	17.5	503
	Average	35.2	30.5	24.7	24.5	19.8	17.8	18.8	533
	1	41.0	34.7	29.2	25.8	21.4	20.1	17.0	600
70	2	44.5	36.2	29.5	25.8	20.8	20.3	17.0	603
	3	43.0	34.1	27.9	26.6	_	19.2	19.7	610
	Average	42.8	35.0	28.9	26.1	21.1	19.9	17.9	604
	1	47.4	37.8	30.6	27.5	21.8	21.0	17.1	634
80	2	44.7	38.2	32.1	29.8	24.2	23.4	18.0	679
	Avergae	46.1	38.0	31.4	28.7	23.0	22.2	17.6	656

¹⁾ Fumigation was conducted at 50, 60, 70 and 80 g/m³ for 24 hours at 15℃ with 54% loading.

 $^{^{2)}}$ 15 cm \times 15 cm \times 40 cm in size.

³⁾ Average moisture content.

Table 2. Mortality data for the pine wood nematode infesting red pine lumber¹⁾ fumigated²⁾ with methyl bromide.

Deser	D1:		sefore fumig	ation	6–7 days after fumigation					
Dose (g/m³)	Repli- cate	Moisture content (%) ³⁾	_		Treatment	Weight of lumber (g)	No. of nematode /100 g	Survivor (%)		
50	1	19.6	288.2	6,200	Fumigation Cont.	277.6 278.7	0.4 3,660	0 59.0		
60	1	20.0	102.1	10,200	Fumigation Cont.	243.4 122.6	0.4 20,147	0 197.5		
00	2	17.5 144.2 22,200		Fumigation Cont.	285.6 142.7	3.5 7,288	0 32.8			
	1	18.0	80.2	8,700	Fumigation Cont.	160.1 81.4	0 35,258	0 405.3		
70	2	2 17.0 160.5 5,400		Fumigation Cont.	191.2	16.1 732	0.3 13.6			
	3	19.7	142.3	34,000	Fumigation Cont.	283.7 141.5	0 19,647	0 57.8		
0.0	1	17.1	-	_	Fumigation Cont.	142.3 143.5	0 16,446	0		
80	2	19.6	288.2	6,200	Fumigation Cont.	281.0 278.7	0 3,660	0.0 59.0		

 $^{^{1)}}$ 15 cm \times 15 cm \times 40 cm in size.

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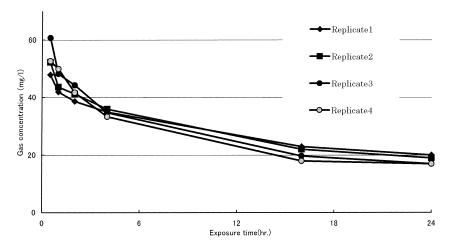


Fig. 1. Progressive gas concentration for New Zealand radiator pine board fumigated with methyl bromide at 48 g/m^3 for 24 hours at $25-34^{\circ}\text{C}$ under tarpaulin.

Table 3 shows CT products from tarpaulin fumigation. The CT product of 620–680 mg·h/l were attained at 48 g/m³ for 24 hours at 26.5–33.5°C with 50–70% loading of New Zealand radiator pine (1.2–4 cm×9–14 cm×90 cm) and 883–1,067 mg·h/l were also attained at 80 g/m³ for 24 hours at 15°C with 25–50% loading of Japanese red pine (3–15 cm×15 cm×100 cm), which were fulfilled minimum CT product of 500 mg·h/l at 48 g/m³ and 700 mg·h/l at 80 g/m³ for a complete moralities of the pine wood nematode, respec-

 $^{^{2)}}$ Fumigation was conducted at 50, 60, 70 and 80 g/m 3 for 24 hours at 15°C with 54% loading.

³⁾ Average moisture content.

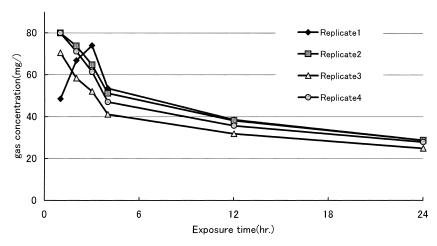


Fig. 2. Progressive gas concentration for Japanese red pine board fumigated with methyl bromide at 80 g/m^3 for 24 hours at 15°C under tarpaulin.

Table 3. CT products in methyl bromide fumigation of pine wood lumber and board under the tarpaulin.

Wood material		Size (cm)	No. of test wood	Moisture content (%)	Loading (%)	Temp. ³⁾ (°C)	Dose (g/m³)	Exposure (h)	CT product (g·h/m³)	Minimum CT product ⁴⁾	
New Zealand radiator pine ¹⁾	1	$\begin{array}{c} 4 \times 14 \times 90 \\ 4 \times 9 \times 90 \end{array}$	20 252	16.0	50	26.5	48	24	679		
	2	$\begin{array}{c} 4 \times 14 \times 90 \\ 4 \times 9 \times 90 \end{array}$	20 252	16.0	50	27.2	48	24	680	500	
	3	$4 \times 9 \times 90$	379	16.0	70	27.3	48	24	653		
	4	1.2×12×90	774	12.3	50	33.5	48	24	620	-	
Japanese red pine ²⁾	1	15×15×100 3×15×100	18 20	31.3 24.3	25	15.2	80	24	1,044		
	2	15×15×100 3×15×100	35 50	31.3 24.3	50	14.8	80	24	1,067	-	
	3	15×15×100 3×15×100	25 99	22.5 15.3	52	15.0	80	24	883	700	
	4	15×15×100 3×15×100	25 99	22.5 15.3	52	15.2	80	24	1,010	-	

 $^{^{1)}}$ Fumigation was conducted under tarpauline $\,$ with circulation fan at ambient temperature.

tively. The CT product of $883-1,067~\text{mg}\cdot\text{h/l}$ fumigated at $80~\text{g/m}^3$ for 24 hours at 15°C with 50-52% loading of 100~cm length of Japanese red pines in tarpaulin fumigation were lower than those of $637-679~\text{mg}\cdot\text{h/l}$ fumigated at $80~\text{g/m}^3$ for 24 hours at 15°C with 54% loading of 40 cm length of Japanese red pines in fumigation box in Table 1 and higher moisture contents and longer size of wood packing materials might cause lower MB gas

²⁾ Fumigation was conducted under tarpauline in the warehouse.

³⁾ Average temperature in tarpaulin.

⁴⁾ Minimum CT product for complete mortalities of the pine wood nematode at different temperatures (Soma *et al.*, 2003).

sorption.

These gas concentrations and CT products in Table 3 showed that the pine wood nematode would be killed completely if no gas-leakage was observed on tarpaulin fumigation.

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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

4. 梱包材を高収容量で臭化メチルくん蒸した場合における CT 値及び殺線虫効果

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マツノザイセンチュウが寄生した含水率 $17\sim20\%$ の赤松角材 $(15~{\rm cm}\times15~{\rm cm}\times40~{\rm cm})$ を 100~l のくん 蒸箱に入れ、収容率 54%, 臭化メチル 50, 60, 70 及び $80~{\rm g/m}^3$ を投薬して $15^{\circ}{\rm C}{\rm C}$ $24~{\rm Bill}$ くん蒸した。平均 CT 値は、 $50~{\rm g/m}^3$ では $455~{\rm mg\cdot h/l}$, $60~{\rm g/m}^3$ では $533~{\rm mg\cdot h/l}$, $70~{\rm g/m}^3$ では $604~{\rm mg\cdot h/l}$, $80~{\rm g/m}^3$ では $656~{\rm mg\cdot h/l}$ で、線虫が完全に殺虫されたのは $80~{\rm g/m}^3$ のみであり、このときの CT 値は $656~{\rm mg\cdot h/l}$ であった。これは、含水量が低い梱包包材を高い収容量でくん蒸する場合、完全殺線虫には高い薬量が必要であることを示している。高い収容量で天幕くん蒸し

た場合の CT 値は、ニュージランドラジエーター松板 材 $(1.2 \sim 4~{\rm cm} \times 9 \sim 15~{\rm cm} \times 90~{\rm cm})$ を収容率 $50~{\rm D}$ び 70%で $48~{\rm g/m}^3$, $26.5 \sim 33.5 {\rm C}$ で $24~{\rm Hfll}$ くん蒸した 場合 $620 \sim 680~{\rm mg}\cdot{\rm h}/l$ で、マツノザイセンチュウの 殺虫に必要な $500~{\rm mg}\cdot{\rm h}/l$ を大きく上回った。また、国産の赤松板材 $(3 \sim 15~{\rm cm} \times 15~{\rm cm} \times 100~{\rm cm})$ を収容率 $25~{\rm D}$ び 50%で $80~{\rm g/m}^3$, $15{\rm C}$ で $24~{\rm Hfll}$ くん蒸した場合 $883 \sim 1,067~{\rm mg}\cdot{\rm h}/l$ を上回った。天幕くん蒸であってもガスの漏洩がなければ、線虫は完全に殺虫されることが判明した。