
An Evaluation of Dazomet Incorporation Methods on Soilborne Organisms and Pine Seedling Production in Southern Nurseries

Stephen W. Fraedrich and L. David Dwinell, 320 Green Street, Southern Research Station, USDA Forest Service, Athens, GA 30602.

ABSTRACT: *The use of dazomet as a fall and spring fumigant for pine seedling production and control of soilborne pests was evaluated at two southern nurseries. Dazomet was applied at low (280-325 kg/ha) and high (493-560 kg/ha) rates and incorporated with a rototiller or spading machine. Comparisons were made with methyl bromide/chloropicrin (MBC) fumigation and nonfumigated control treatments. Dazomet incorporation method had no effect on seedling density at either nursery, and often did not affect seedling morphological characteristics. At the Georgia (GA) nursery, seedling density and morphological characteristics did not differ among fumigant treatments except in the spring study area where shoot weight was greater in the MBC treatment than the dazomet or nonfumigated control treatments. In the study area at the North Carolina (NC) nursery, seedling density was greater in the high-rate dazomet treatment than the nonfumigated control. Seedlings were generally larger in MBC and dazomet treatments than the control. Seedling density and morphological characteristics did not differ among fumigation treatments in the spring study area. Fumigation with MBC or dazomet generally reduced the percentage of roots with Pythium and Fusarium spp. compared to controls at the GA nursery and the fall fumigation area in the NC nursery. Plant parasitic nematodes were found infrequently at both nurseries and did not differ among treatments. Nutsedge (Cyperus spp.) was the major problem at the GA nursery and was effectively controlled only with MBC. Compared to the MBC treatment, the abundance of soilborne fungi and the association of certain types of Trichoderma with roots was often lower in the dazomet treatments. The importance of these differences for long term seedling production and management of soilborne diseases is not known at this time. South. J. Appl. For. 27(1):41-51.*

Key Words: Pest management, fumigation, pine, forest-tree nurseries, seedling production, methyl bromide, dazomet.

Southern nursery managers have relied on soil fumigation with methyl bromide for more than 40 yr to control soilborne diseases, insects, nematodes, and weeds. In 1993, approximately 92% of southern nursery managers routinely used methyl bromide for pest control (Fraedrich and Smith 1994). Methyl bromide has been identified as an ozone depleting agent, and its use as a soil fumigant is to be phased out by 2005 under the U.S. Clean Air Act and the Montreal Protocol (Environmental Protection Agency 1999).

Dazomet (Mylone®, Basamid®) has been used with success in some western and northern forest-tree nurseries; however, the fumigant has not gained widespread acceptance in southern nurseries. Dazomet had been tried by 40% of southern nursery managers surveyed, but most indicated dissatisfaction with this chemical (Fraedrich and Smith 1994). Some managers who have tested dazomet believe it provides poorer weed and disease control than methyl bromide fumigation, and may reduce seedling quality (Fraedrich and Smith 1994). However, information

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is lacking on procedures used to test dazomet in the individual trials. Various factors may influence the efficacy of dazomet including soil conditions, rate of application, incorporation method, activation of the material, and time between application and sowing. Incorporation method affects the distribution of dazomet in soil (Kelpsas and Campbell 1994, Juzwik et al. 1997), and application rate and incorporation method together can greatly influence the control of soilborne pathogenic fungi (Juzwik et al. 1999). Compared to fumigation with methyl bromide/chloropicrin, fumigation with dazomet requires a longer aeration time before seed sowing. Therefore, a narrower window of opportunity is available for fumigation in the late winter and spring than during the fall months. The purpose of this study was to examine factors that may affect the performance of dazomet in southern pine nurseries. The specific objectives of the study were to evaluate dazomet rate, method of incorporation, and season of fumigation on pine seedling production at two nurseries that differed in geographic location and soil type. We also evaluated the effect of dazomet on plant parasitic nematodes, weeds, and fungi that were present in soil and on roots of pine seedlings.

Materials and Methods

Study Sites

Studies were conducted at the Flint River Nursery (Georgia Forestry Commission) near Byromville, Georgia (GA) and the Edwards Nursery (North Carolina Division of Forest Resources) near Morganton, North Carolina (NC). The soil at the GA nursery is a loamy sand and classified in the Eustis soil series. The soil at the NC nursery is a loam and classified in the Congaree soil series. Both nurseries fumigate routinely with methyl bromide/chloropicrin (MBC) prior to pine seedling production. At the GA nursery, the study was conducted in a field removed from production in the early 1990s because of problems thought to be caused by nematodes and fungi. At the NC nursery, the study was conducted in two adjacent fields that were to be fumigated before operational production of seedlings.

Experimental Design and Application of Treatments

Two study areas were established at each nursery. Each study area was either a fall or spring fumigation consisting of two rates of dazomet, a spring MBC fumigation treatment, and a nonfumigated control. Two tillage implements, a rototiller and a spading machine, were also evaluated for incorporation of dazomet.

The experimental design was a split plot with replication by means of blocks. The basic split-plot design for one area is illustrated in Figure 1. The whole plots compared tillage implements, while the subplots compared fumigation treatments. Whole plots were 195 m long and 4.9 m wide (three beds) at the GA nursery, and each was divided into subplots that were 24.4 m long. At the NC nursery, whole plots were 183 m by 4.9 m (three beds), and each subplot was 22.9 m long. Each tillage implement and fumigation treatment combination was replicated three times at the NC nursery and four times at the GA nursery.

Block 1	Tiller	MBC	DL	DH	DL	C	DL
	Spader	C	DL	DH	MBC	C	DL
Block 2	Tiller	C	DL	C	MBC	DH	DH
	Spader	DL	DL	C	MBC	DH	MBC
Block 3	Spader	DL	DH	MBC	C	C	DH
	Tiller	MBC	DL	C	C	DH	DH
Block 4	Spader	MBC	DH	DL	DL	C	C
	Tiller	C	DH	MBC	C	C	DL

Figure 1. The design for the fall study area at the Georgia nursery. Tillage implements (spader or tiller) were used over the entire plot length. Treatments applied to subplots were: MBC—methyl bromide/chloropicrin; DH—dazomet, high rate; DL—dazomet, low rate; and C—control. Hatch-filled areas are untreated buffers. The study design was similar at the North Carolina nursery, but only three blocks were used in each study area.

Dazomet was applied at the GA nursery on October 11, 1996 and March 11, 1997, in the fall and spring study areas, respectively. At the NC nursery, dazomet was applied on October 29, 1996 in the fall study area and March 3 1, 1997 in the spring study area. Soil temperatures at 10 cm depth were recorded between 1200 and 1400 hr during the dazomet application. Temperatures at the GA nursery were approximately 20°C at the time of the fall and spring applications. At the NC nursery, the soil temperature during the fall application was 20.5°C, and 15.5°C for the spring application.

Dazomet was applied at rates of 325 kg/ha and 493 kg/ha to randomly selected subplots during fall 1996 at the GA nursery. For the spring application at the GA nursery, and for both the fall and spring dazomet applications at the NC nursery, low and high rates of dazomet were 280 and 560 kg/ha, respectively. Dazomet was applied over the soil surface with a Gandy® drop spreader. Dazomet was incorporated using a Ferguson® rototiller with 20 cm tines, or a Gramegna® spading machine with a maximum operating depth of 30 cm. Each tillage implement was operated continuously across the whole plot in each block. The ground speed for the spading machine during the incorporation of dazomet was 1.9–2.4 kilometers per hour (kph), and 1.9–2.7 kph for the rototiller. The power take off (PTO) for the rototiller and spader were operated at 540 revolutions per minute. The surface of all plots was sealed with a bed roller. Recommendations of the manufacturer (BASF®) were followed for soil irrigation after incorporation of dazomet (Pennington 1995). Amounts of water applied were based on nursery estimates of the output rate of their overhead irrigation system and duration of watering. At the GA nursery, approximately 2.5 cm of water was applied to all plots immediately after incorporation; 1.9 cm on the second day, 1.2 cm on the third day, 0.6 cm on the fourth day, and 0.2 cm on the fifth day. On the sixth and seventh days after application of dazomet, sufficient water was applied to maintain surface moisture. At the NC nursery the water regime was modified to prevent puddling and runoff. On the day of application, 2.5 cm of water was applied to plots. On the second through the fourth days only 0.6 cm of water was applied daily. On the fifth day, 0.2 cm was applied, and on the sixth and seventh days, sufficient water was applied to maintain surface moisture. In both nurseries, fields were harrowed after the seventh day, and plots were permitted to air for at least 2 wk before seed sowing. MBC (67% methyl bromide and 33% chloropicrin) was applied to

randomly selected subplots in all study areas on March 19, 1997 at the GA nursery and on May 6, 1997 at the NC nursery. The MBC fumigation was applied at 392 kg/ha using the standard practices for each nursery.

Soil samples were collected from all subplots treated with dazomet before seed sowing, and a radish seed germination test was performed to determine the presence of residual fumigant. Soil was placed in Mason® jars, moistened, and 25 radish seeds were placed on the soil surface. The jars were sealed and germination was evaluated after 5-10 days. Germination was not inhibited in any of the treatments.

All plots were operationally sown with slash pine (*Pinus elliotii* Engelm. var. *elliotii*) seeds at the GA nursery on May 5, 1997 and with loblolly pine (*P. taeda* L.) seeds at the NC nursery on May 12, 1997. All other chemical applications (e.g., herbicides and fungicides) and cultural practices (e.g., fertilization, watering, and undercutting) were uniformly applied to all study areas. Seedlings were top-pruned at the NC nursery but not at the GA nursery. Seedbed density (number of seedlings per m²) and morphological characteristics were evaluated in the center bed of each subplot.

Seedling Bed Density and Morphological Characteristics

Three permanent sample plots, each 0.3 m x 1.2 m, were randomly established in each subplot immediately after sowing to monitor and evaluate seedling bed density. After the first seedling counts on May 21, 1997 at the GA nursery, seedlings were lost on the margins of some beds because of soil erosion. Therefore, all sample plots at the GA nursery were subsequently reduced in size to 0.3 m x 0.6 m and located in the center of beds. Live and dead seedlings were counted at 10–20 day intervals during the first 6-8 wk after sowing. A final assessment was made in December 1997 (NC) and January 1998 (GA).

Seedling root collar diameter (RCD), shoot height (root collar to the apex of the shoot), and shoot and root dry weight were evaluated at the end of the growing season. Seedlings were lifted from three 0.3 m x 0.6 m sample plots selected at random over the length of each subplot. Fifteen seedlings were randomly selected and evaluated from each sample plot (45 per subplot). Seedlings were dried at 80°C for 48-72 hr, and root and shoot weights were determined.

Soilborne Mycoflora

On all subplots treated with the rototiller, the colony forming units (cfu's) of *Fusarium* and *Pythium* spp. and "total fungi" associated with soils were assessed in June (GA) and in July (NC) of 1997. Since only subplots on the rototilled plots were assessed for these variables, the design for this portion of the study was simplified to a randomized complete block with four fumigation treatments and three (NC) or four (GA) blocks. Composite soil samples consisting of 8-10 soil cores (2.5 cm wide and 15.25 cm depth) were obtained on each subplot. Soil was mixed thoroughly and appropriate soil dilutions were prepared. The soil dilutions were plated on potato dextrose agar amended with tergitol, ampicillin, and rifampicin (PDATAR) (Kannwischer et al. 1994) for general fungal counts; PARP medium (Singleton et al. 1992) for *Pythium* spp.; and Nash-Snyder's agar medium (Nelson et al.

1983) for *Fusarium* spp. Five plates of each medium were used for each subplot, and on each plate was placed 1 ml of the soil dilution. Plates were incubated in growth chambers at 25°C. Plates with PARP medium were evaluated after 48-72 hr. Plates with PDATAR and Nash-Snyder's media were evaluated after 5-7 days.

Association of Fungi With Roots

Root systems were evaluated just prior to lifting for the presence of *Pythium*, *Fusarium*, *Trichoderma*, and *Gliocladium* spp. A sample of 3-4 seedlings was collected at each of three random locations on each subplot (10 seedlings total). Roots were rinsed with tap water, surface sterilized in a 0.5% sodium hypochlorite solution for 1 min., rinsed three times with sterile water, and blotted dry with sterile paper towels. Roots were cut into sections 5 to 8 cm long and plated on Komada's (Nelson et al. 1983), PDATAR and PARP media. Three root sections for each seedling were placed on one plate of each of the three agar media (nine sections/seedling).

Root samples on agar media were incubated in growth chambers at 25°C. Roots on the PARP medium were evaluated after 48-72 hr, the others after 5-7 days. Roots were scored for the presence of *Fusarium*, *Pythium*, *Trichoderma*, *Gliocladium*, and other common fungi. *Trichoderma* spp. were classified to type based on their cultural morphology. Selected isolates of each type of *Trichoderma* spp. were grown on malt extract agar and identified to species aggregate based on the keys of Rifai (1969) and Bissett (1991).

Selected *Fusarium* isolates were transferred to KC1 and carnation leaf agar, and when necessary potato dextrose agar. Isolates were identified to species using the taxonomic key of Nelson et al. (1983). *Pythium* isolates were grown on corn meal agar and in grass/water culture, and identified using the taxonomic keys of Middleton (1943), and Van der Plaats-Niterink (1981).

Plant Parasitic Nematode Assessments

Composite soil samples were collected as previously described for all subplots in June or July, and again at lifting to evaluate for plant parasitic nematodes. Nematodes were extracted using standard procedures and identified to genus by the Cooperative Extension Services at the University of Georgia.

Weed Evaluations

Nutsedge (*Cyperus* spp.) plants were counted in mid-June at the GA nursery on three randomly selected 0.3 m x 1.2 m sample plots in each subplot. Other weeds were rarely encountered in treatment plots at the GA nursery. All weeds were counted on the entire subplots in the fall and spring study areas at the NC nursery just before the nursery initiated its operational herbicide program.

Analysis of Data

The study was analyzed as a split-plot design with replication by means of blocks for all variables except for the soilborne mycoflora (i.e., *Fusarium* spp., *Pythium* spp. and "total fungi"). Tillage implement was the whole plot factor and fumigation treatment was the subplot factor for the split

plot design. For soilborne mycoflora variables, only the fumigation treatments on the rototilled plots were assessed, and therefore the design was simplified to a randomized complete block with four treatments. An analysis of variance (ANOVA) was conducted for all variables, and differences among means were evaluated with Tukey's honest significant difference (HSD) procedure (Cochran and Cox 1968, Milliken and Johnson 1984). Nutsedge data at the GA nursery was often not normally distributed, and a nonparametric split-plot analysis was performed based on ranking the data within each block before proceeding with the analysis (Conover 1980, SAS® Procedures Guide 1990). For all variables, significance levels were set at the $P = 0.1$ level in the ANOVA and subsequent mean separation tests.

Results

Georgia Nursery

Seedling Bed Density, Morphological Characteristics and Mortality

No differences were detected among fumigation treatments, tillage implements or their interactions for bed densities, root-collar diameter, shoot height, and shoot weight in the fall study area (Table 1). Although there was a significant interaction of tillage implement and fumigation treatment for root weight ($P = 0.0984$), no differences could be detected among means. In the spring study area, the shoot weight was greater in the MBC treatment than in the dazomet and control treatments. Although the treatment effect for shoot height was significant ($P = 0.0930$), no differences among means were detected. No other differences were observed for seedling morphological characteristics among tillage implements, fumigation treatments or their interaction.

Mortality was rarely observed in sample plots in either the fall or spring study areas (Table 2), and when observed, was due to a variety of factors including insects, birds, possible soilborne and seedborne fungi, and field equipment.

Table 1. Slash pine seedling density, morphological characteristics and nutsedge density for fumigation treatments in fall and spring study areas at a Georgia nursery.

Study area	Treatment	Seedling&?	RCD (mm)	Height (cm)	Root wt	Shoot wt	Nutsedge
					(g)	(shoots/m ²)*
Fall	Control	317 a [†]	4.5 a	29.3 a	0.998 a	3.60 a	57 a
	Dazomet (325 kg/ha)	306 a	4.4 a	29.4 a	0.914 a	3.64 a	23 ab
	Dazomet (493 kg/ha)	327 a	4.2 a	29.3 a	0.924 a	3.50 a	13 bc
	MBC ^{††}	317 a	4.6 a	30.0 a	0.967 a	3.97 a	3 c
	Treatment ($P > F$ value)	0.2563	0.3784	0.94	0.5126	0.1516	<0.0001
	TMSD [‡]	24.5	0.44	3.22	0.152	0.494	
Spring	Control	323 a	4.2 a	29.5 a	0.873 a	3.19 a	98 a
	Dazomet (280 kg/ha)	314 a	4.1 a	29.0 a	0.860 a	3.14 a	71 a
	Dazomet (560 kg/ha)	320 a	4.3 a	29.7 a	0.835 a	3.34 a	36 a
	MBC	327 a	4.5 a	33.6 a	0.851 a	4.06 b	4 b
	Treatment ($P > F$ value)	0.4804	0.1813	0.0930	0.8868	0.0056	0.0007
	TMSD	22.4	0.43	4.73	0.121	0.614	—

* Data was ranked, and nonparametric split-plot analysis was conducted on ranked data. Actual means are presented in table.

[†] Means followed by the same letter do not differ significantly according to Tukey's HSD procedure ($P \leq 0.10$). Means based on 8 replications per treatment.

^{††} MBC = methyl bromide/chloropicrin.

[‡] TMSD = Tukey's minimum significant difference.

Soilborne Mycoflora

Pythium spp. were absent or detected only at low levels in soil fumigated with MBC or dazomet in the fall and spring study areas, but frequently, no differences could be detected between controls and other treatments (Table 3). More cfu's of *Fusarium* spp. were found in the nonfumigated control soil than in all other treatments in the fall study area. No differences were observed between the dazomet treatments and the MBC treatment. In the spring study area, the cfu's of *Fusarium* spp. were greater in the control than in the MBC and low-rate dazomet treatments but not the high rate dazomet treatment. In the fall study area, the total fungal cfu's were less in the dazomet-treated soil than in the MBC-treated soil. In addition, fewer fungal cfu's were found in soil treated with the low rate of dazomet compared to the control. The cfu's for all fungi were greater in MBC-treated soil compared to all other treatments in the spring study area.

Fungi Associated with Roots

The association of *Pythium* spp. with roots was greater in the nonfumigated controls than in other fumigation treatments for both study areas (Table 4). In addition, a significant tillage implement by fumigation treatment interaction ($P = 0.0312$) was observed for *Pythium* spp. in the fall study area. For the control treatments, the isolation of *Pythium* spp. from roots did not differ between rototiller ($\bar{x} = 24.1\%$) and spader ($\bar{x} = 11.5\%$). However, the isolation of *Pythium* spp. in the control treated with the rototiller differed from all other fumigation treatments treated with the rototiller (range of means: 0-6.6%) or the spader (range of means: 0.1-3.3%). *Fusarium* spp. were frequently isolated from roots of seedlings in all treatments, but were more common on roots in the controls than in the high-rate dazomet and the MBC treatments in both study areas.

The *Pythium* spp. most commonly isolated from roots was *Pythium irregulare* Buisman. *Fusarium oxysporum* Schlecht. was isolated from 82% of root segments; *F. proliferatum*

Table 2. Summary of observed mortality by fumigation treatment and sample date at Georgia and North Carolina nurseries.

Nursery	Study area	Treatment	Dead seedlings/m ² on observation date:*				
			I [†]	II	III	IV	V
GA	Fall	Control	0.0	0.0	0.4	0.0	0.2
		Dazomet (325 kg/ha)	0.0	0.2	0.2	0.0	0.0
		Dazomet (493 kg/ha)	0.0	0.4	0.6	0.0	0.2
		MBC ^{††}	0.0	0.2	1.6	0.0	0.6
	Spring	Control	0.1	0.0	0.4	0.2	0.0
		Dazomet (280 kg/ha)	0.0	0.0	0.6	0.2	0.0
		Dazomet (560 kg/ha)	0.0	0.0	2.5	0.2	0.0
		MBC	0.0	0.0	0.8	0.0	0.0
NC	Fall	Control	0.0	8.5	1.9	1.2	0.0
		Dazomet (280 kg/ha)	0.0	6.6	2.2	0.4	0.0
		Dazomet (560 kg/ha)	0.0	10.0	3.4	0.6	0.0
		MBC	0.0	8.5	1.6	0.6	0.0
	Spring	Control	0.0	16.1	7.0	1.1	0.0
		Dazomet (280 kg/ha)	0.0	12.2	5.9	0.9	0.0
		Dazomet (560 kg/ha)	0.0	12.2	4.8	0.9	0.0
		MBC	0.0	9.4	5.8	0.6	0.0

* Mortality due to various factors including insects, birds, chemical injury, sunscald, seedborne and soilborne fungi, and routine nursery operations such as undercutting and lateral pruning.

[†] Observation dates at the GA were: I-5/21/97, II-5/29/97, III-6/13/97, IV-7/28/97, and V-1/8/98, and observation dates at the NC were: I-5127197, II-619197, III- 6/18/97, IV- 7/14/97 and V-1213197.

^{††} MBC = methyl bromide/chloropicrin.

(Matsushima) Nirenberg and *F. solani* (Mart.) Sacc. were isolated from only 12% and 8% of the root segments, respectively.

Two types of *Trichoderma* were recognized from roots at the GA nursery. Type-A isolates were blue-green with aerial mycelium that was often clumped. Sterile hook-shaped hyphae were noted at the margins of the colonies. Type-B isolates were often dark green, or had a yellow-green coloration that became a darker green with age. Generally, aerial mycelium in Type-B isolates was continuous across petri dishes and not in clumps as in Type-A isolates.

No differences were observed among fumigation treatments for the presence of *Trichoderma* Type-A isolates on roots in either study area. Type-B isolates were more

commonly associated with roots in the MBC treatment than other treatments in the fall and spring study areas. However, in the spring study area, the Type-B isolates were more commonly associated with roots in the high-rate dazomet treatment than in the control. In the fall study area, the Type-B isolates also occurred with greater frequency ($P = 0.0014$) in plots treated with the spader ($\bar{x} = 45.6\%$) than the rototiller ($\bar{x} = 38.3\%$). *Gliocladium* spp. were recovered more frequently from roots in the control and MBC treatments than from roots in the dazomet treatments. *Gliocladium* spp. were also isolated with greater frequency ($P = 0.0365$) from roots in the rototiller ($\bar{x} = 13.1\%$) than spader ($\bar{x} = 5.4\%$) treatment in the fall study area.

The Type-B isolates were predominantly *T. harzianum* Rifai, although other unidentified species also were present.

Table 3. Mean colony forming units (cfu) of *Pythium* spp., *Fusarium* spp., and "total fungi" from soil of fumigation treatments in fall and spring study areas at a Georgia nursery.

Study area	Treatment	Mean cfu/g* soil		
		<i>Pythium</i> spp.	<i>Fusarium</i> spp.	Total fungi
Fall	Control	42 a [†]	1,553 b	22,303 bc
	Dazomet (325 kg/ha)	0 a	475 a	6,649 a
	Dazomet (493 kg/ha)	11 a	446 a	9,885 ab
	MBC ^{††}	0 a	224 a	36,793 c
	Treatment (P > F value)	0.2431	0.0069	0.0013
	TMSD [§]	66	975	15,598
Spring	Control	117 b	2,561 b	34,619 a
	Dazomet (280 kg/ha)	0 a	1,147 a	17,390 a
	Dazomet (560 kg/ha)	11 a	1,388 ab	22,638 a
	MBC	32 ab	481 a	138,489 b
	Treatment (P > F value)	0.0440	0.0089	<0.0001
	TMSD	100	1,265	36,142

* cfu/g = colony forming units per gram.

[†] Means within study area followed by the same letter do not differ significantly according to Tukey's HSD procedure ($P \leq 0.10$). Means based on 4 replications per treatment.

^{††} MBC = methyl bromide/chloropicrin.

[§] TMSD = Tukey's minimum significant difference.

Table 4. Mean percentage of slash pine roots from which *Pythium*, *Fusarium*, *Trichoderma* and *Gliocladium* spp. were isolated for fumigation treatments in fall and spring study areas at a Georgia nursery.

Study area	Treatment	Percentage of roots*				
		<i>Pythium</i> spp.	<i>Fusarium</i> spp. (Type-A)	<i>Trichoderma</i> (Type-B)		<i>Gliocladium</i> spp.
Fall	Control	18 a ^{††}	95 a	37 a	22 a	15 a
	Dazomet (325 kg/ha)	4 b	67 b	28 a	29 a	5 b
	Dazomet (493 kg/ha)	2 b	58 bc	24 a	30 a	2 b
	MBC [§]	2 b	52 c	33 a	87 b	15 a
	Treatment ($P > F$ value)	<0.0001	<0.0001	0.3150	-0.0001	0.0017
	TMSD		12	18	12	8
Spring	Control	27 a	92 a	54 a	16 a	15 a
	Dazomet (280 kg/ha)	6 b	78 ab	45 a	33 ab	19 ab
	Dazomet (560 kg/ha)	1 b	65 b	50 a	45 b	18 ab
	MBC	2 b	65 b	60 a	68 c	34 b
	Treatment ($P > F$ value)	<0.0001	0.0100	0.5209	<0.0001	0.0940
	TMSD	10	20	27	18	19

* Assessment based on isolation from three root segments per seedling, 10 seedlings per subplot (80 seedlings per treatment).

† Type-A isolates were primarily *T. hamatum* (Bon) Bain; Type-B isolates were predominantly *T. harzianum* Rifai.

†† Means followed by the same letter do not differ significantly according to Tukey's HSD procedure ($P \leq 0.10$). Means based on 8 replications per treatment.

§ MBC = methyl bromide/chloropicrin.

|| TMSD = Tukey's minimum significant difference.

The Type-A isolates were primarily *T. hamatum* (Bon) Bain. The *Gliocladium* isolates were primarily *G. viride* Matr.

Plant Parasitic Nematodes

No differences were detected among tillage implements, fumigation treatments or their interaction for plant parasitic nematodes in the fall and spring study areas. Plant parasitic nematodes occurred at low levels, and means among fumigation treatments ranged from 1.2 to 28.2 per 100 cc of soil. Ring nematodes (*Criconeimoides* spp.) were the most common plant parasitic nematode, although root knot nematodes (*Meloidogyne* spp.) occasionally occurred in large numbers (up to 120 nematodes/100 cc soil) in the end-of-year samples. Also present were the lesion (*Pratylenchus* spp.), stubby root (*Trichodorus* spp.), spiral (*Helicotylenchus* spp.) and cystoid (*Meloidodera* spp.) nematodes.

Weed Assessments

Purple nutsedge (*Cyperus rotundus* L.), and to a lesser extent yellow nutsedge (*C. esculentus* L.), were the principal weeds not controlled by herbicides used in the nursery's operational program. MBC virtually eliminated nutsedge and was the only fumigation treatment to consistently reduce nutsedge populations (Table 1). Although dazomet provided some control of nutsedge, control was not as consistent or as great as that provided by MBC. Nutsedge plants appeared to increase throughout the summer in dazomet-treated and nonfumigated soils; no increase was observed in the MBC-treated subplots.

North Carolina Nursery

Seedling Bed Density, Morphological Characteristics, and Mortality

Seedbed density was greater in the high-rate dazomet treatment than in nonfumigated control in the fall study area

(Table 5). No other significant differences in bed densities were observed among fumigation treatments, tillage implements, or implement by fumigation treatment interaction in either the fall or spring study areas. Root collar diameter, shoot height, and root and shoot weights were often greater for seedlings in MBC and dazomet treatments than in the nonfumigated control in the fall study area. No differences in morphological characteristics occurred between the MBC and dazomet treatments. Also in the fall study area, shoot height was greater ($P = 0.0309$) in plots treated with the rototiller ($\bar{x} = 24.6$ cm) than the spader ($\bar{x} = 23.2$ cm). In addition, root weight was greater ($P = 0.0421$) in plots treated with the rototiller ($\bar{x} = 0.938$ g) compared to the spader ($\bar{x} = 0.771$ g). In the spring study area, a significant tillage implement by fumigation treatment interaction was noted for root weights ($P = 0.0444$); however, significant differences could not be detected among fumigation treatments within or among tillage implements. No other differences were observed for seedling morphological characteristics in the fall or spring study area.

Seedling mortality was observed in all treatments during evaluations on June 9, 1997 (Table 2). However, based on symptoms, we believe that these losses were due to sunscald or chemical injury. Seedlings frequently had localized stem lesions at or above the ground level, but we did not observe necrosis typical of that associated with damping-off even after seedlings were moist-chambered for 2 days. Similar mortality was also observed throughout operational areas of the nursery. The presence of dead seedlings in seed beds decreased with later observation dates.

Soilborne Mycoflora

Pythium spp. were rarely isolated from soil of any treatment in either the fall or spring study areas (Table 6). The cfu's of *Fusarium* spp. were greater in the nonfumigated

Table 5. Loblolly pine seedling density, morphological characteristics and weed density for fumigation treatments in fall and spring study areas at a North Carolina nursery.

Study area	Treatment	Seedlings/m ²	RCD (mm)	Height (cm)	Root wt (g)	Shoot wt	Weeds (no./m ²)
Fall	Control	223 a*	4.0 a	22.5 a	0.723 a	0.73 a
	Dazomet (280 kg/ha)	250 ab	4.6 b	24.6 b	0.861 ab	3.06 b	0.17 b
	Dazomet (560 kg/ha)	260 b	4.6 b	24.6 b	0.925 b	3.35 b	0.15 b
	MBC [†]	230 ab	4.7 b	24.1 ab	0.908 b	3.12 b	0.29 b
	Treatment (P > F value)	0.0300	0.0035	0.0220	0.0398	0.0157	0.0010
	TMSD ^{‡‡}	31	0.4	1.6	0.170	0.70	0.30
Spring	Control	241 a	4.8 a	22.5 a	0.867 a	2.57 a	0.14 a
	Dazomet (280 kg/ha)	226 a	5.0 a	22.2 a	0.951 a	2.70 a	0.29 a
	Dazomet (560 kg/ha)	227 a	4.8 a	23.0 a	0.903 a	2.81 a	0.19 a
	MBC	238 a	4.8 a	22.5 a	0.879 a	2.57 a	0.14 a
	Treatment (P > F value)	0.7596	0.6917	0.7433	0.1540	0.4282	0.5400
	TMSD	45	0.4	1.6	0.093	0.43	0.28

* Means followed by the same letter do not differ significantly according to Tukey's HSD procedure ($P \leq 0.10$). Means based on 6 replications per treatment.

[†] MBC = methyl bromide/chloropicrin.

^{‡‡} TMSD = Tukey's minimum significant difference.

control than in the dazomet and MBC treatments in the fall study area. The cfu's of all fungi were greater in MBC-treated soil than in soil of all other treatments in the fall study area. In the spring study area, no differences were observed among treatments for cfu's of *Fusarium* spp. or total fungi.

Fungi Associated with Roots

The effect of tillage implements, or the interaction of tillage implements and fumigation treatments were not significant for any of the fungi associated with roots in either the fall or spring study areas. However, the effect of fumigation treatments was significant for all variables in the fall study area (Table 7). Compared to the nonfumigated control treatment, MBC and dazomet treatments reduced the association of *Pythium* and *Fusarium* spp. with roots. In the spring study area, no differences could be detected among treatments for the association of either *Fusarium* or *Pythium* spp. with roots.

Fusarium oxysporum was isolated from 93% of all root segments; *F. solani* and *F. proliferatum* were also isolated from roots, but infrequently. The *Pythium* isolates were not keyed to species.

Two types of *Trichoderma* were also isolated from roots at the NC nursery. The colony morphology of the Type-A isolates was similar to those found at the GA nursery, but colonies did not have the bluish coloration and were often pale green. Sterile, spiral-shaped hyphae were noted on the margins of the colonies. Type-B isolates were similar to those found at the GA nursery.

In the fall study area, Type-A isolates were associated more commonly with roots in the MBC and control treatments than the dazomet treatments. In contrast, Type-B isolates were obtained more frequently from roots in the high-rate dazomet treatment than the MBC or control treatments. *Gliocladium* spp. were less frequently associated with

Table 6. Mean colony forming units (cfu) of *Pythium* spp., *Fusarium* spp., and "total fungi" from soil of fumigation treatments in spring and fall study areas at a North Carolina nursery.

Study area	Treatment	Mean cfu/g* soil		
		<i>Pythium</i> spp.	<i>Fusarium</i> spp.	Total fungi
Fall	Control	15 a [†]	4,485 a	29,979 a
	Dazomet (280 kg/ha)	0 a	415 b	38,533 a
	Dazomet (560 kg/ha)	15 a	143b	19,728 a
	MBC ^{‡‡}	0 a	473 b	110,734 b
	Treatment (P > F value)	0.5957	0.0001	0.0011
	TMSD ^{‡‡}	42	1,028	40,594
Spring	Control	0	586 a	23,125 a
	Dazomet (280 kg/ha)	0	505 a	13,521 a
	Dazomet (560 kg/ha)	0	197a	12,425 a
	MBC	0	629 a	21,410 a
	Treatment (P > F value)		0.8378	0.7452
	TMSD		1,412	32,211

* cfu/g = colony forming units per gram.

Means within study area followed by the same letter do not differ significantly according to Tukey's HSD procedure ($P \leq 0.10$). Means based on 3 replications per treatment.

^{‡‡} MBC = methyl bromide/chloropicrin.

[‡] TMSD = Tukey's minimum significant difference.

Table 7. Mean percentage of loblolly pine roots from which *Pythium*, *Fusarium*, *Trichoderma* and *Gliocladium* spp. were isolated for fumigation treatments in fall and spring study areas at a North Carolina nursery.

Study area	Treatment	Percentage of roots*				
		<i>Pythium</i> spp.	<i>Fusarium</i> spp.	<i>Trichoderma</i> †		<i>Gliocladium</i> spp.
				(Type-A)	(Type-B)	
Fall	Control	70 a ^{††}	93 a	64 a	5c	26 a
	Dazomet (280 kg/ha)	11 b	55 b	37 b	37 ab	5b
	Dazomet (560 kg/ha)	15 b	41 b	34 b	58 a	3b
	MBC [§]	32 b	48 b	84 a	20 bc	13b
	Treatment (P > F value)	0.0005	0.0065	0.0002	0.0011	0.0004
	TMSD	27	33	22	25	10
Spring	Control	15 a	72 a	62 a	28 a	32 a
	Dazomet (280 kg/ha)	21 a	64 a	52 a	24 a	26 a
	Dazomet (560 kg/ha)	6 a	44 a	42 a	35 a	11 a
	MBC	17a	64 a	47 a	36 a	34 a
	Treatment (P > F value)	0.5820	0.4596	0.6000	0.8959	0.1213
	TMSD	28	44	38	44	25

* Assessment based on isolations from three root segments per seedling, 10 seedlings per subplot (60 seedlings per treatment).

† Type-A isolates were predominantly *T. spirale* Bissett; Type-B isolates included *T. harzianum* Rifai, although other unidentified species also were present.

†† Means followed by the same letter do not differ significantly according to Tukey's HSD procedure ($P \leq 0.10$). Means based on 6 replications per treatment.

§ MBC = methyl bromide/chloropicrin.

|| TMSD = Tukey's minimum significant difference.

roots in the MBC or dazomet treatments than the nonfumigated control. In the spring study area, no differences occurred among fumigation treatments for the association of *Trichoderma* and *Gliocladium* spp. with roots.

The Type-A isolates were predominantly *T. spirale* Bissett, although *T. hamatum* was also found. The Type-B isolates included *T. harzianum*, although other unidentified species also were present. A mix of *Gliocladium virens* (J. Miller, Giddens, Fostare) von Arx, Beih. and *G. viride* was observed at this nursery.

Plant Parasitic Nematodes

No differences were detected among tillage implements, fumigation treatments, or their interaction for plant parasitic nematodes in the fall and spring study areas. Plant parasitic nematodes were seldom observed, and means among fumigation treatments ranged from 0 to 7.3 nematodes per 100 cc of soil. Lesion, stubby root, stunt (*Tylenchorhynchus* spp.) and spiral nematodes were found at this nursery.

Weed Assessments

Fumigation with MBC or dazomet reduced the number of weeds compared to the nonfumigated control in the fall study area (Table 5). No significant differences were observed among treatments in the spring study area. The herbicide program at the NC nursery provided effective control of most weeds in study areas.

Discussion

Fumigation with dazomet has been beneficial for the production of conifer seedlings and control of soilborne pests in some western nurseries (Alspach 1989, Campbell and Kelpsas 1988, Hildebrand 1991, McElroy 1986, Tanaka et al. 1986, Tkacz and Ramirez 1988), and nurseries in the northern states (Borkenhagen 1994, Enebak et al. 1990a.). In this

study, we did not encounter major problems with the use of dazomet, and with respect to seedling size and production, the results were often similar to fumigation with MBC.

One potential problem with dazomet is that it did not control nutsedge as well as MBC. A general concern of some nursery managers has been that dazomet does not control weeds as effectively as methyl bromide (Fraedrich and Smith 1994). Dazomet did not control weeds in one study conducted in southern nurseries, but the rates of dazomet were 336 kg/ha or less (Carey 1994). Chapman (1992) also noted poor control of nutsedge and several other weed species with dazomet at rates of 336-392 kg/ha. According to Roman et al. (1994), commercially acceptable control of susceptible weed species can be achieved when dazomet is applied at rates above 392 kg/ha, and nutsedge control can be achieved when rates are above 448 kg/ha and conditions are optimum. In our study, dazomet provided some control of weeds at both locations. Nonetheless, even at rates as great as 560 kg/ha, dazomet was not consistently as effective as MBC for nutsedge control. If dazomet is to be used for nutsedge control, additional efforts will be necessary to better define the optimal use conditions.

Stunted seedlings and losses in seedling production associated with the use of dazomet were not found in this study although some nursery managers have reported such problems (Fraedrich and Smith 1994). As with any fumigant, application techniques and soil conditions are critical for maximum effectiveness. The activation and efficacy of dazomet are directly dependent on moisture, and the application of sufficient moisture is a key factor in the use of this chemical (Munnecke and Martin 1964). Without adequate moisture, activation can be delayed and extended, and concentrations of methylisothiocyanate may not reach critical levels for control of soilborne pests. The soil aeration period

after fumigation is also critical to permit the evacuation of fumigant from soil. An inadequate aeration period may adversely affect seed germination and seedling quality. In one study, pine seeds had to be resown on dazomet-treated plots because the residual toxic effects of the fumigant killed seedlings from the first sowing (Ridge and Theodorou 1972). Practices and procedures that do not fully activate dazomet or allow a sufficient aeration period before sowing may account for some of the variable results in the use of this chemical.

The method of dazomet incorporation has been shown to influence the distribution of the chemical and control of soilborne pathogens (Kelpsas and Campbell 1994, Juzwik et al. 1997, Juzwik et al. 1999). Certain types of tillage implements can incorporate dazomet deeper into soil and provide a better distribution of the chemical. In this study, no consistent differences were found between the rototiller and spading machine. Seedling production and control of potential soilborne pests were generally comparable with either method of dazomet incorporation.

Fumigation with dazomet or MBC had definite effects on the soilborne mycoflora and the fungi associated with roots. Except in the spring study area at the NC nursery, dazomet generally reduced cfu's of *Fusarium* spp. in soil and the association of *Fusarium* and *Pythium* spp. with the roots. It is not certain if the *Fusarium* and *Pythium* spp. associated with the roots were pathogenic, but we rarely found evidence of root disease or damping-off. The effect of dazomet on potentially pathogenic soilborne fungi has been highly variable among reported studies and trials in nurseries. Barnard et al. (1994) found that dazomet can provide effective control of *Macrophomina phaseolina*, *Fusarium* spp., and *Pythium* spp. in southern nurseries. Studies elsewhere in the United States have found that dazomet reduces populations of *Fusarium* and *Pythium* spp. to levels similar to methyl bromide (Hildebrand 1991, Tanaka, et al. 1986, Tkacz and Ramirez 1988). In other studies, the effect of dazomet on soilborne *Fusarium* spp. was negligible or very limited (Hildebrand and Dinkel 1988, Campbell and Kelpsas 1988; Enebak et al. 1990b). Differences in chemical rates, application techniques, site conditions, and study protocol may explain some of the variability among studies.

Danielson and Davey (1969) found that *Trichoderma* spp. and certain other saprophytic fungi increased soon after fumigation with methyl bromide. Although we often noted this type of response with MBC, we rarely observed a similar type response with dazomet. The total fungal cfu's following dazomet fumigation were either similar to, or less than that found in nonfumigated soils. The association of certain *Trichoderma* spp. with roots was often significantly less in the dazomet treatments than the MBC treatment. One notable exception was in the fall study area at the NC nursery where *Trichoderma* Type-B isolates were more commonly associated with roots in the high-rate dazomet treatment than the MBC treatment. *Trichoderma* and *Gliocladium* spp. are well known for their ability to act as microbial antagonists of plant pathogenic fungi and many are recognized as biological control agents

(Hjeljord and Tronsmo 1998). Species of these fungi are also believed to directly promote plant growth even in the absence of plant diseases (Bailey and Lumsden 1998). At this time the relationship of southern pine seedling quality to the presence of specific *Trichoderma* and *Gliocladium* spp. in soil and on roots is not known.

Fumigation with either dazomet or MBC provided benefits in terms of seedling production and growth in one study area, but in other study areas, the benefits of fumigation were not as clear. Although fumigation provided a seedling growth response in the fall study area at the NC nursery, the seedlings in the nonfumigated control treatment were not stunted. An analysis of these seedlings by grade indicated that fumigation increased the percentage of grade 1 seedlings (RCD>4.8 mm) but had no significant effect on the percentage of cull seedlings (RCD<3.2 mm) or plantable seedlings (RCD>3.2 mm). The mean percentage of grade 1 seedlings in the dazomet and MBC treatments ranged from 38 to 43%, but only 14% were grade 1 in the nonfumigated control. The mean percentage of cull seedlings ranged from 5.6 to 8.5% among treatments. The reasons for the seedling growth response with fumigation are not understood. Weeds were not a problem at the NC nursery, and soilborne diseases were not observed. Growth increases following fumigation have been noted in other studies where there have not been serious soilborne diseases. Abiotic factors believed to account for such plant growth responses include increased availability of ammonium nitrogen, manganese, and phosphorus following fumigation (Alexander 1967, Ingestad and Molin 1960, Rovira 1976). Changes in the beneficial soil microbiota following fumigation are also believed to affect seedling growth (Ingestad and Nilsson 1964).

Weed control has been a major reason for fumigation in the past. Advances in herbicides over the last 25-30 yr have raised questions about the need to fumigate routinely for weed control in southern nurseries (Chapman 1992). South and Gjerstad (1980) showed that herbicides can control most broadleaf and annual weed problems in southern nurseries. They indicated that fumigation was warranted for difficult-to-control weeds such as nutsedge when populations were abundant, but once fumigated, nutsedge can be kept under control with spot applications of herbicides such as glyphosate (South 1984, South and Gjerstad 1980). The herbicide EPTC also can be used to control nutsedge (Hodges 1960, Rowan 1961). In addition, South (1979) outlined a comprehensive weed management program that relies on herbicides and a variety of nonchemical weed-control techniques and less intensively on fumigation.

Disease outbreaks or major seedling losses were not observed at either nursery. Post-germination losses due to diseases in nonfumigated soil also have not been noted in other recently conducted studies in southern nurseries (Carey 1996, Carey 2000, Barnard et al. 1997, Fraedrich and Dwinell 1996). The possibility exists that the lack of nematode and disease problems in the nonfumigated areas in these studies may be related to operational fumigation from years past, and disease potential over the long term may be underestimated

by current studies. Recently, a disease problem associated with a plant-parasitic nematode was found in one nursery field that had been in continuous production for several years (Fraedrich and Cram 2001). Nonetheless, disease problems have been the exception in recent studies, and the lack of diseases underscores the need for development of more comprehensive integrated pest management (IPM) programs that can better assess the necessity for fumigation in any particular field. Currently, many fields are routinely fumigated after every pine crop or every other pine crop, and disease control is often cited as a reason for the fumigation (Cram and Fraedrich 1996, Fraedrich and Smith 1994). Nurseries can differ dramatically in soil type, management practices, and many other factors that affect disease development. A better understanding of the pathogens and factors that contribute to soilborne diseases in southern pine nurseries could lead to development of more effective IPM programs and overall cost reductions for management of pests.

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