Chemical Alternatives to Methyl Bromide for Nematode Control under Vineyard Replant Conditions

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Abstract: Vineyard replant disorder is a disease of unknown etiology, currently controlled by methyl bromide fumigation. While not all the components of the disease are known, plant-parasitic nematode densities are often high in vineyard replant soils. Alternatives to the broadly effective general biocide, methyl bromide, are needed for vineyards replanted after January 2005, when import and manufacture of methyl bromide was banned except for exempted quarantine uses and approved critical uses. Two field trials were conducted in vineyards that had been planted to own-rooted Thompson Seedless grapes for 70 to 85 years and were known to support populations of plant-parasitic nematodes. In a randomized block design with five or six replicates per trial, shank-injected and/or drip-applied propargyl bromide (>207 kg ha^{-1}), iodomethane + chloropicrin (50:50, >269 kg ha^{-1} drip-applied), and 1,3-dichloropropene + chloropicrin (InLine, 468 L ha^{-1}) provided control of plant-parasitic nematodes throughout the first growing season similar to control achieved with methyl bromide (507 kg ha^{-1}). Chloropicrin (448 kg ha^{-1}) provided less nematode control than methyl bromide, but significantly greater control than untreated. An experimental drip formulation of sodium azide (Agrizide, 336 kg ha^{-1}) was insufficient to control nematodes under vineyard replant conditions. Although vine growth in the treated plots was generally greater than in the untreated plots, growth was greater with methyl bromide than with the alternative chemicals. Alternatives to methyl bromide for nematode control in sandy loam soils were documented, but acceptable alternatives for the management of the complex vineyard replant disorder are more elusive.

Key words: drip fumigation, fallow, nematode, resistance

Success in replanting a vineyard requires the elimination or minimization of physical or biological constraints found in the field. “Replant disorder” is a general term for the lack of vigor in a newly replanted vineyard as compared to vines planted in “nonvineyard” soil. Multiple factors, which vary between sites, can contribute to replant disorder, resulting in stunted plants, leaf chlorosis, and uneven growth across the vineyard (McKenry 1999, Westphal et al. 2002). Replant disorder appears to be more severe in areas where old grape roots are abundant and has been associated with plant-parasitic nematodes, phylloxera, and soilborne diseases (Deal et al. 1972, McKenry 1992). Vines and trees have extensive root systems that cannot be completely removed when orchards and vineyards are removed. Live grape roots can survive for more than eight years after a vineyard has been removed and can serve as a reservoir of inocula for nematodes and pathogens (McKenry and Buzo 1996). Even low populations of *Meloidogyne* spp. Chitwood (root-knot nematode), *Tylenchulus semipenetrans* Cobb (citrus nematode), *Pratylenchus vulnus* Allen and Jensen (lesion nematode), and *Xiphinema index* Thorne and Allen (dagger nematode) in the soil can result in extensive damage if grapes are replanted into a field within three years after perennials were grown (McKenry 1992).

The current practice in California for replanting grapes into a vineyard with a known soilborne pest problem is to fumigate with either methyl bromide or 1,3-dichloropropene (1,3-D). Both fumigants are effective against nematodes and can provide kill of old roots to a depth of 1.5 to 2 m (McKenry and Buzo 1996). In 1993, methyl bromide was designated as a primary ozone-depleting compound by the parties of the Montreal Protocol. Under this international treaty, methyl bromide importation and production levels were reduced by 25% beginning in 1999, followed by a 50% reduction in 2001, and a 70% reduction in 2003. All reductions were taken from the 1991 baseline. Production and importation of methyl bromide in the United States was required to cease by 1 Jan 2005, with exemptions for quarantine/preshipment and critical uses. Use of 1,3-D is limited in California by township caps, which restrict the amount of 1,3-D that can be applied in each township in a calendar year (Cal DPR 2001).

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The search for alternatives to methyl bromide has included old chemicals not currently in wide use, alternative application methods for commercially available compounds, and evaluation of new materials. Iodomethane, also called methyl iodide, has previously been tested against wireworms and for use in postharvest storage applications (Lehman 1942, Burditt et al. 1963). Recent tests using iodomethane as a soil fumigant have shown broad spectrum efficacy against pathogens, nematodes, and weeds similar to that obtained for methyl bromide (Becker et al. 1998, Hutchinson et al. 1999, Ohr et al. 1996, Schneider et al. 2004, Zhang et al. 1997). Propargyl bromide, a compound patented by Dow Chemical in 1957, was an effective broad spectrum soil fumigant, but it was explosive (Ajwa et al. 2002a, Hutchinson et al. 2004, Yates and Gan 1997). Although earlier trials with drip-applied nematicides provided mixed results, recent trials of 1,3-D, chloropicrin, and metam sodium applied through drip-irrigation systems have demonstrated good results for some annual and perennial crops (Ajwa et al. 2002a, Roberts et al. 1988, Schneider et al. 2003, Westerdahl et al. 1993, 2003). Sodium azide in a granular formulation was not effective as a nematicide under field conditions, but new liquid formulations have been used in a number of recent studies in annual cropping systems (Gerik et al. 2002, Kelley and Rodriguez-Kabana 1978, Rodriguez-Kabana et al. 2004).

In commercial vineyards, several plant-parasitic nematode genera often occur at the same time. When vineyards are replanted, resistant rootstocks can be useful, but rarely include resistance against all the genera that are present (Anwar et al. 2002, McKenry et al. 2001a,b). While rootstocks such as Harmony or Freedom can be very effective against root-knot nematodes, suppressing the root-knot nematode population in the soil environment can create an opportunity for other nematode genera to increase.

Although each management strategy has its strengths and weaknesses, combining strategies has the potential to provide greater nematode control in situations where a single management practice alone might not be sufficient for the multiyear life of perennial crops. The purpose of the current research is to determine the effect of potential methyl bromide alternatives under vineyard replant conditions on plant-parasitic nematode populations at planting and at the end of the first growing season, as well as on weed and crop growth in the first growing season. The alternatives examined are experimental chemicals, alternative methods of applying fumigants, and resistant rootstocks.

Materials and Methods

2001 Trial. An 85-year-old, plant-parasitic nematode-infested vineyard of *Vitis vinifera* L. cv. Thompson Seedless located at the USDA Parlier, CA, research station was selected for this field trial. Soil type was a Hanford fine sandy loam. Vines were removed in Oct 2000 and the vineyard was ripped to a depth of 90 cm in two directions. The experimental design was a randomized complete block with 10 treatments and 5 replications. Each plot was 11 m wide and 15 m long. All treatments were applied in April 2001. The shank-injected treatments were commercially applied by Tri-Cal (Hollister, CA) using a deep-shank fumigation rig. Shanks were set 1.5 m apart and 56 cm deep. The fumigant was injected and 0.03-mm polyethylene tarp laid in a single operation.

The drip-applied treatments were made in 75 mm of water over 16 hr through buried (25 cm deep) thin-walled drip-irrigation tubes (0.78 L h^{-1}) with emitters spaced 30 cm apart. The amount of water was estimated as the amount needed to penetrate to a soil depth of 120 cm based on soil texture and soil water content at the time of fumigation. Drip tubes were spaced 60 cm apart to obtain a broadcast treatment over the entire plot. That contrasts with bed treatments, commonly used in crops such as strawberry, in which only the shaped and tarped plant bed is treated (Ajwa and Trout 2004). The fumigant was delivered from pressurized cylinders, through a manifold, and into the drip lines using the method described by Ajwa et al. (2002b). All drip treatments received a cap of either metam sodium (243 L ha^{-1} of 42% metam sodium [Vapam HL]) in 12 cm water or 12 cm water alone applied through microspray sprinklers to seal the soil surface and, in the case of the metam sodium caps, to control weeds.

There were 10 treatments: (1) untreated control; (2) shank-injected methyl bromide (99.5% methyl bromide, 0.5% chloropicrin) at 507 kg ha^{-1}; (3) shank-injected iodomethane:chloropicrin (50:50) at 515 kg ha^{-1} (Midas, Arysta Corp., San Francisco, CA); (4) shank-injected propargyl bromide at 221 kg ha^{-1}; (5) microspray-applied Vapam HL at 243 L ha^{-1} (42% metam sodium); (6) drip-applied 1,3-D:chloropicrin at 468 L ha^{-1} (InLine, 61% 1,3-D, 33% chloropicrin + emulsifier) followed by a metam sodium cap; (7) drip-applied chloropicrin at 448 kg ha^{-1} followed by a metam sodium cap; (8) drip-applied iodomethane: chloropicrin (50:50) at 448 kg ha^{-1} (Midas) followed by a water cap; (9) drip-applied propargyl bromide at 207 kg ha^{-1} followed by a water cap; and (10) drip-applied sodium azide at 336 kg ha^{-1} (Agrizide) followed by a water cap. In order to determine the contribution of the metam sodium cap to nematode control, a stand-alone metam sodium cap treatment was included at 243 L ha^{-1}. The 243 L ha^{-1} rate is appropriate for use of metam sodium as an herbicide, but it is lower than the 525 to 701 L ha^{-1} rate recommended on the label for perennial replant situations.

In early June 2001, each plot was planted with one row each of own-rooted Thompson Seedless (susceptible to root-knot and citrus nematodes), Merlot on 1103P (moderately resistant to root-knot nematode), or Thompson Seedless on Freedom rootstock (resistant to some root-knot nematodes, susceptible to citrus nematodes) dormant bare-root vines (McKenry et al. 2001b). Vines were spaced 1.8 m in the row and 3.7 m between rows.

Soil samples were collected at planting with an 8-cm diam bucket auger from the center of each plot in 30 cm increments to a depth of 150 cm. Soil samples were thoroughly mixed and a 300 cm^3 subsample removed. Samples were processed using the sieving/Baermann funnel protocol that recovers only those nematodes that could reasonably be assumed to be still viable as plant parasites (Flegg and Hooper 1970). Nematodes were identified and counted under the microscope. Nematode counts were transformed using ln(n+1) to stabilize the variance (Noe 1985). The transformed counts were analyzed using ANOVA and means separated using the Waller–Duncan K-ratio *t*-test option in GLM (version 6; SAS Institute, Inc., Cary, NC). Weeds were removed in mid-July from a strip 50 cm wide and 1.2 m long centered on the middle plant row in each plot. The aboveground weed biomass was air-dried for two weeks and weighed.

In Nov 2001, after one full growing season, soil samples were collected with a 2.5-cm diam sampling tube to a depth of 60 cm. Two cores were collected from each of the central five plants in each row of each plot and composited to obtain one sample for each variety/rootstock combination. These samples were processed using the sieving/sugar flotation/centrifugation protocol with a 500-mesh sieve (25 µm opening) (Jenkins 1964). Nematodes were identified and counted under the microscope and data were analyzed as previously described. In Jan 2002, vines were pruned to two nodes above the graft union or an equivalent position on the own-rooted vines. The prunings were dried at 60°C for seven days and then weighed. The data were analyzed using ANOVA and means separated using the Waller–Duncan K-ratio *t*-test.

In Sept 2003 and Aug 2004, berries were picked and weighed. The data were analyzed using ANOVA and means separated using the Waller–Duncan K-ratio *t*-test.

2003 Trial. A 70-year-old, plant-parasitic nematode-infested vineyard of Thompson Seedless located at the USDA Parlier, CA, research station was selected for the second field trial. Soil type was a Hanford sandy loam. Vines were removed in Sept 2002 and the vineyard was ripped to a depth of 90 cm in two directions. The experimental design was a randomized complete block with seven treatments and six blocks. Each plot was 11 m wide. Shank-injected methyl bromide plots were 39 m long to allow for accurate delivery of the specified amount of chemical. All other plots were 19 m long. There were seven treatments: (1) untreated control; (2) shank-injected methyl bromide (99.5% methyl bromide, 0.5% chloropicrin) at 448 kg ha^{-1}; (3) drip-applied iodomethane:chloropicrin (50:50) at 268 kg ha^{-1} (Midas) followed by a metam sodium cap; (4) drip-applied iodomethane:chloropicrin (50:50) at 336 kg ha^{-1} followed by a metam sodium cap; (5) drip-applied 1,3-D:chloropicrin at 468 L ha^{-1} (InLine, 61% 1,3-D, 33% chloropicrin + emulsifier) followed by a metam sodium cap; (6) drip-applied Agrizide (sodium azide in Agri-50) at 336 kg ha^{-1} delivered through drip tape placed at 5 cm depth (shallow depth); and (7) drip-applied Agrizide (sodium azide in Agri-50) at 336 kg ha^{-1} delivered through drip tape placed at 25 cm depth (standard depth). The shank treatment was commercially applied by Tri-Cal in Oct 2002 as previously described. The drip-applied treatments were applied in Nov 2002 in 95 mm water over 16 hr through drip-irrigation tubes (1.06 L h^{-1}) with emitters spaced 30 cm apart. The drip tape was buried at 5 cm depth for the shallow Agrizide treatment and at 25 cm for all other treatments. All drip treatments received a cap of either metam sodium or water. Drip-fumigation and microspray cap protocols were as described in the 2001 trial.

In April 2003, each plot was planted with one row each of own-rooted Thompson Seedless, Merlot on 1103P, or Autumn Royal on Harmony (resistant to some root-knot nematodes, susceptible to citrus nematodes) dormant bareroot vines (McKenry et al. 2001b). Vines were spaced 2.4 m in the row and 3.7 m between rows.

Soil samples were collected at planting, processed, nematodes identified and counted, and data analyzed as described for the 2001 trial. Weeds were removed six weeks after vine planting (six months after treatment), dried and weighed as described in the 2001 trial. In Nov 2003, after the first growing season, soil samples were collected, processed, and analyzed as described in the 2001 trial. In Jan 2004, vines were pruned to two nodes above the graft union, prunings weighed, and data analyzed as previously described.

Results

In the 2001 trial, populations of the citrus nematode were present in all five soil zones to a depth of 150 cm in the untreated control plots at the time of planting (Table 1). Nematode populations in the methyl bromide control were virtually undetectable to a soil depth of 150 cm. All chemical treatments, except one, resulted in nematode populations of less than 4 nematodes per 100 cm^3 soil at all soil depths. The drip-applied sodium azide treatment had slightly, but not statistically significant, higher nematode populations, compared to the methyl bromide treatment at the time of planting. The metam sodium cap treatment applied through microsprays had high citrus nematode populations that were not significantly different from the untreated control.

Weed biomass data from plots treated with metam sodium caps in two of the five replicates suggested that application problems had compromised the data. All weed data from these two replicates were removed from further analyses. Data from the remaining three replicates were analyzed as previously described. Methyl bromide-treated plots had no weeds three months after treatment, while the metam sodium herbicide treatment had very low counts (Table 1). Plots treated with shank-injected Midas or propargyl bromide, or any drip application with a metam sodium cap, had more weed biomass than the methyl bromide control, but significantly less than the untreated control. Weed control in the sodium azide-treated plots was not significantly different from either the methyl bromide or the untreated control. The drip treatments with water caps had weed levels not significantly different from the untreated plots.

After one growing season, both citrus nematode and root-knot nematode were found in the untreated control plots (Table 2). Nematodes were essentially below detectable levels on all three variety/rootstock combinations for plots treated with methyl bromide, shank-injected and drip-applied Midas, shank-injected and drip-applied propargyl bromide, and drip-applied InLine. Drip-applied chloropicrin-treated plots had slightly higher root-knot nematode populations on own-rooted Thompson Seedless and Merlot on 1103P than in methyl bromide-treated plots, but were still an order of magnitude lower than in the untreated control plots. Drip-applied sodium azide-treated plots had higher nematode populations than the other chemical treatments, but lower than the untreated control after one growing season. *Meloidogyne* spp. populations in Thompson Seedless and 1103P rootstock rows in sodium azide-treated plots were not significantly different from population levels in the untreated control plots. *Tylenchulus semipenetrans* population levels in sodium azide-treated plots were significantly lower than popula-

Table 1 *Tylenchulus semipenetrans*, citrus nematode, populations at planting per 100 cm³ soil in a vineyard replant trial planted in 2001. Values are the antilogs of the means of (ln(n+1)) for five replications. Weed biomass measured three months after treatment, mean of three replications.

Treatment	Rate a.i. (ha⁻¹)	*Tylenchulus semipenetrans* 0–30 cm	30–60 cm	60–90 cm	90–120 cm	120–150 cm	Weeds (g)
Untreated	—	122.5 a[a]	73.7 b	50.9 b	22.4 b	4.2 c	537.4 a
Methyl bromide	507 kg	0.0 c	0.0 c	0.0 d	0.0 c	1.2 c	0.0 c
Shank Midas	515 kg	3.5 bc	1.3 c	1.5 d	0.0 c	0.0 c	38.5 c
Shank propargyl bromide	221 kg	0.0 c	0.0 c	1.2 d	0.0 c	0.0 c	146.9 bc
Metam sodium cap	243 L	845.6 a	1332.5 a	1556.2 a	431.4 a	66.0 b	9.7 c
Drip InLine + metam sodium cap	468 L	1.6 bc	1.6 c	1.4 d	0.0 c	0.0 c	80.5 c
Drip chloropicrin + metam sodium cap	448 kg	0.0 c	0.0 c	2.0 d	2.3 c	2.0 c	66.9 c
Drip Midas + water cap	448 kg	1.2 c	0.0 c	0.0 d	1.3 c	0.0 c	620.2 a
Drip propargyl bromide + water cap	207 kg	0.0 c	0.0 c	1.4 d	0.0 c	1.2 c	492.4 ab
Drip sodium azide + water cap	336 kg	7.8 bc	0.0 c	3.6 cd	4.3 bc	5.2 c	303.2 abc

[a]Values for each depth followed by the same letter are not significantly different at the $p = 0.05$ level.

Table 2 *Meloidogyne* sp. and *Tylenchulus semipenetrans* populations in the top 60 cm soil per 100 cm³ soil sampled after one growing season, mean of five replications, in a vineyard replant trial planted in 2001. Statistical analyses conducted on log-transformed (ln(n+1)) data. Data presented are the antilogs of the means.

Treatment	*Meloidogyne* sp. (/100 cm³ soil)			*Tylenchulus semipenetrans* (/100 cm³ soil)		
	Thompson Seedless	Thompson Seedless/ Freedom	Merlot/ 1103P	Thompson Seedless	Thompson Seedless/ Freedom	Merlot/ 1103P
Untreated	323.5 a[a]	11.1 a	25.3 a	120.6 a	30.8 a	49.9 a
Methyl bromide	0.0 c	0.0 c	0.0 c	0.0 c	0.0 c	0.0 c
Shank Midas	0.0 c	0.0 c	0.1 c	0.1 c	0.0 c	1.5 c
Shank propargyl bromide	1.5 bc	0.0 c	0.3 c	0.0 c	0.7 bc	0.0 c
Metam sodium cap	290.2 a	3.5 b	34.2 a	156.6 a	32.1 a	15.9 ab
Drip InLine + metam sodium cap	0.0 c	0.1 c	0.0 c	0.0 c	0.0 c	0.0 c
Drip chloropicrin + metam sodium cap	7.6 b	0.3 c	5.6 b	2.3 bc	0.4 c	0.7 c
Drip Midas + water cap	0.9 bc	0.0 c	0.0 c	0.0 c	0.0 c	0.0 c
Drip propargyl bromide + water cap	0.0 c	0.1 c	0.0 c	0.2 c	0.0 c	0.0 c
Drip sodium azide + water cap	63.8 a	1.1 bc	11.6 ab	9.1 b	3.0 bc	3.8 bc

[a]Means for each nematode genus followed by the same letter are not significantly different at the $p = 0.05$ level.

tions in untreated plots for all three variety/rootstock combinations. Citrus nematode population levels on Freedom and 1103P rootstocks in sodium azide-treated plots were not significantly different from population levels in the methyl bromide plots. Plots treated with the herbicidal rate of metam sodium supported nematode populations similar to the untreated control plots. Freedom and 1103P rootstocks supported lower populations of both root-knot nematode and citrus nematode than did own-rooted Thompson Seedless.

Thompson Seedless both on its own root and on Freedom showed significant differences in pruning weights between plants grown in untreated and methyl bromide-treated plots (Table 3). Own-rooted Thompson Seedless vines from propargyl bromide plots were not significantly different in size from vines grown in methyl bromide-treated plots nor were Thompson Seedless on Freedom vines grown in InLine-treated plots. Vines grown in sodium azide-treated plots were not significantly different in size from vines grown in untreated plots for any variety/rootstock combination. While there was a numeric difference in vine size for the Merlot vines on 1103P rootstock grown in methyl bromide-treated plots and those grown in untreated plots, the difference was not statistically significant when all 10 treatments were compared. The largest vines were found in the shank-injected propargyl bromide and methyl bromide-treated plots and the smallest vines were in the shank-injected Midas-treated plots. When the analysis was limited to only the methyl bromide and untreated plots, the pruning weights of the Merlot on 1103P were significantly greater in plots treated with methyl bromide than in untreated plots. Some burning of leaf edges and chlorosis was observed throughout the first growing season in vines grown in plots treated with either shank-injected or drip-applied Midas.

Table 3 Pruning dry weights collected in Jan 2002 from trial planted in 2001.

Treatment	Thompson Seedless dry wt (g)	Thompson on Freedom dry wt (g)	Merlot on 1103P dry wt (g)
Untreated	4.5 e[a]	7.8 e	10.4 abc
Methyl bromide	20.3 a	39.4 a	16.1 ab
Shank Midas	14.4 b	25.6 bc	8.8 c
Shank propargyl bromide	16.8 ab	21.9 bcd	16.3 a
Metam sodium cap	14.2 bc	25.0 bc	11.9 abc
Drip InLine + metam sodium cap	14.0 bc	31.7 ab	13.8 abc
Drip chloropicrin + metam sodium cap	13.5 bc	24.0 bcd	14.3 abc
Drip Midas + water cap	14.1 bc	25.0 bc	10.1 bc
Drip propargyl bromide + water cap	16.3 abc	19.6 cd	12.7 abc
Drip sodium azide + water cap	6.7 de	17.7 cde	9.5 c

[a]Means for each variety/rootstock combination followed by the same letter are not significantly different at the $p = 0.05$ level.

Yield per vine differed significantly among treatments for own-rooted Thompson Seedless vines in the first year of harvest (Table 4). Shank-injected Midas and all drip-applied treatments, except sodium azide, resulted in yields comparable to methyl bromide. Vines in the untreated plots yielded 40% of the fruit harvested in methyl bromide-treated plots. There were no significant differences for any other variety/rootstock combination in the first year. There were no significant differences in yield for any treatment in the second harvest year.

In the 2003 trial, citrus nematodes were present at the time of planting in the untreated plots in all five soil layers to a depth of 150 cm (Table 5). Nematode populations in all other treatments were below detectable levels with the exception of the shallow application of Agrizide, where low populations, not significantly different from methyl bromide, were detected.

Qualitative observations made of weed control in Feb 2003 indicated that plots treated with metam sodium caps

Table 4 Yield in 2003 and 2004 from trial planted in 2001.

Treatment	2003 yield (kg/vine)			2004 yield (kg/vine)		
	Thompson Seedless	Thompson on Freedom	Merlot on 1103P	Thompson Seedless	Thompson on Freedom	Merlot on 1103P
Untreated	7.56 d[a]	18.90	4.86	20.79	19.42	11.32
Methyl bromide	18.61 a	21.75	7.37	19.43	24.21	11.35
Shank Midas	16.06 a	16.78	6.15	20.61	18.57	10.32
Shank propargyl bromide	11.92 c	19.25	6.30	20.59	21.53	13.66
Metam sodium cap	11.87 c	18.32	5.70	20.72	22.58	11.73
Drip InLine + metam sodium cap	15.67 ab	17.63	6.92	21.18	21.66	10.85
Drip chloropicrin + metam sodium cap	15.71 ab	21.85	5.22	21.56	22.90	11.23
Drip Midas + water cap	17.42 a	17.70	6.90	23.26	23.72	12.27
Drip propargyl bromide + water cap	17.45 a	19.85	7.89	24.26	21.92	11.58
Drip sodium azide + water cap	9.82 cd	20.62	5.06	19.79	20.27	11.15

[a]Means for each cultivar/rootstock combination followed by the same letter are not significantly different at the $p = 0.05$ level.

applied through microsprays were nearly free of all weeds. Methyl bromide-treated plots contained a few *Malva* spp. plants. The sodium azide delivered in shallow-placed drip tape provided good weed control. The sodium azide delivered through standard-depth tapes provided good weed control immediately above the drip tape, but a row of weeds grew vigorously between the drip tapes. The untreated plots had a solid cover of weeds. Three months later, when weed biomass was measured, large differences were no longer apparent (Table 5). Weed control was greatest in plots that received a cap of metam sodium applied through microsprays. The methyl bromide-treated plots had significant weed pressure from *Malva* spp. The least weed control was observed in plots treated with sodium azide.

At the end of the first growing season, both root-knot and citrus nematode were found in the untreated plots (Table 6). The standard application of Agrizide gave better control of root-knot nematode than did the shallow application, but neither gave control comparable to methyl bromide except in plots planted with the Harmony rootstock. Methyl bromide, both rates of Midas, and InLine resulted in populations of both root-knot and citrus nematode not significantly different from zero for all variety/rootstock combinations.

Significant differences in pruning weights between plants grown in methyl bromide-treated plots and those grown in untreated plots were documented for all variety/rootstock combinations (Table 7). The largest vines were grown in plots treated with methyl bromide for all variety/rootstock combinations. The smallest vines were grown in either the untreated plots or plots treated with sodium azide. In all cases, the pruning weights of vines grown in plots treated with azide were not significantly different from the pruning weights of vines grown in the untreated plots. Vines grown in plots treated with Midas or InLine were intermediate in size, with the exception of Merlot on 1103P grown in plots treated with the higher rate of Midas, which were not significantly different in size from vines grown in methyl bromide-treated plots. No phytotoxicity was observed in the plots treated with Midas in the 2003 trial.

Table 5 *Tylenchulus semipenetrans* populations per 100 cm^3 soil sampled at planting March 2003, mean of six replications. Statistical analyses conducted on log-transformed (ln(n+1)) data. Data presented are the antilogs of the means. Aboveground weed dry weight (g) collected in May 2003.

		***Tylenchulus semipenetrans* (/100 cm^3 soil)**					
Treatment	**Rate (ha^{-1})**	**0-30 cm**	**30-60 cm**	**60-90 cm**	**90-120 cm**	**120-150 cm**	**Weeds (g)**
Untreated control	—	60.3 a[a]	78.6 a	181.6 a	78.3 a	10.0 a	363.7 ab
Methyl bromide	448 kg	0.0 b	0.0 b	0.0 b	0.0 b	0.0 b	335.2 ab
Midas (low) + metam sodium cap	269 kg	0.0 b	0.0 b	0.0 b	0.0 b	0.0 b	306.3 ab
Midas (high) + metam sodium cap	336 kg	0.0 b	0.0 b	0.0 b	0.0 b	0.0 b	103.3 b
InLine + metam sodium cap	468 L	0.0 b	0.0 b	0.0 b	0.0 b	0.0 b	236.7 ab
Sodium azide, shallow placement	336 kg	0.2 b	1.6 b	0.3 b	0.0 b	0.0 b	409.6 a
Sodium azide, standard placement	336 kg	0.0 b	0.0 b	0.0 b	0.0 b	0.0 b	417.2 a

[a]Means for each depth followed by the same letter are not significantly different at the $p = 0.05$ level.

Table 6 *Meloidogyne* sp. and *Tylenchulus semipenetrans* populations in the top 60 cm soil per 100 cm^3 soil sampled after one growing season in Nov 2003, mean of six replications, in a vineyard replant trial planted in 2003. Statistical analyses conducted on log-transformed (ln(n+1)) data. Data presented are the antilogs of the means.

	***Meloidogyne* sp. (/100 cm^3 soil)**			***Tylenchulus semipenetrans* (/100 cm^3 soil)**		
Treatment	**Thompson Seedless**	**Merlot/ 1103P**	**Autumn Royal/ Harmony**	**Thompson Seedless**	**Merlot/ 1103P**	**Autumn Royal/ Harmony**
Untreated control	35.7 a[a]	12.6 a	4.9 a	68.8 a	24.4 a	2.5 a
Methyl bromide	0.0 c	0.0 d	0.0 c	0.0 b	0.0 c	0.0 b
Midas (low)	0.1 c	0.1 d	0.0 c	0.0 b	0.0 c	0.0 b
Midas (high)	0.0 c	0.4 d	0.1 c	0.0 b	0.0 c	0.0 b
InLine	0.2 c	0.0 d	0.0 c	0.0 b	0.0 c	0.0 b
Sodium azide, standard depth	3.7 b	1.6 c	0.0 c	0.0 b	0.9 b	0.0 b
Sodium azide, shallow depth	76.0 a	4.2 b	1.3 b	0.0 b	1.5 b	0.0 b

[a]Means for each nematode genus followed by the same letter are not significantly different at the $p = 0.05$ level.

Table 7 Pruning dry weights collected in Jan 2004 from trial planted in 2003, means of six replications.

Treatment	Thompson Seedless dry wt (g)	Autumn Royal/ Harmony dry wt (g)	Merlot/ 1103P dry wt (g)
Untreated control	47.7 d[a]	54.1 d	68.9 e
Methyl bromide	279.1 a	225.7 a	256.5 a
Midas (low)	155.5 bc	80.2 bcd	148.5 cd
Midas (high)	181.4 b	96.6 bc	223.4 ab
InLine	144.1 c	107.0 b	180.5 bc
Sodium azide, standard depth	44.5 d	60.6 cd	80.3 de
Sodium azide, shallow depth	51.2 d	47.0 d	104.1 de

[a]Means for each variety/rootstock combination followed by the same letter are not significantly different at the $p = 0.05$ level.

Discussion

These field trials were conducted under true vineyard replant conditions by removing vines from commercial vineyards that were more than 70 years old and were known to be infested with plant-parasitic nematodes. The vines were removed and the soil prepared in a manner consistent with commercial practices to ensure that the results in these trials would be representative of results under commercial conditions. Substantial populations of plant-parasitic nematodes were encountered throughout the sampled soil profile to a depth of 150 cm. Potential alternatives to methyl bromide for use in vineyard replant must be evaluated for efficacy throughout the soil profile. As shown by these trials, even populations of root-knot nematode so low as to be virtually nondetectable at planting can build up during the course of the first growing season.

Methyl bromide alternatives vary in their spectrum of activity. Iodomethane, propargyl bromide, sodium azide, and metam sodium have broad spectrum activity under optimal conditions (Becker et al. 1998, Hutchinson et al. 1999, 2004, Lehman 1942, Martin 2003, Ohr et al. 1996, Roberts et al. 1988, Skroch et al. 1977). 1,3-Dichloropropene is used primarily as a nematicide. Chloropicrin is a broad spectrum fumigant, but at the rates most commonly used is primarily a fungicide. Chloropicrin is often combined with other materials such as 1,3-D to provide a broader spectrum of control or with methyl bromide or iodomethane to obtain a synergistic effect (Hutchinson et al. 2000, Munnecke and Van Gundy 1979). Metam sodium can be applied sequentially with other fumigants to enhance weed control (Ajwa and Trout 2004). Chemical rates evaluated in this trial were chosen based on economic considerations, regulatory restrictions, and rates found to be effective in other cropping systems. While higher rates of iodomethane and propargyl bromide, such as molar equivalents to the rate of methyl bromide used in this study, might have been even more effective, cost of iodomethane and regulatory concerns for propargyl bromide coupled with results from other cropping systems led us to evaluate rates with a higher probability of acceptance from regulatory agencies and cost-conscious growers.

Unregistered compounds. Midas (a combination of iodomethane and chloropicrin), propargyl bromide, and sodium azide each demonstrated some efficacy in reducing populations of plant-parasitic nematodes throughout the first growing season under vineyard replant conditions. Although these three compounds are not new, the need for alternatives to methyl bromide has renewed interest in these materials. Midas controlled plant-parasitic nematodes as well as methyl bromide at planting and after one growing season. Midas was effective at controlling nematodes when applied by deep shank-injection or by drip-application. These results concur with results using iodomethane, either alone or in combination with chloropicrin, obtained in other cropping systems (Becker et al. 1998, Hutchinson et al. 1999, Ohr et al. 1996, Schneider et al. 2004). Midas is not currently commercially available, but a registrant has submitted the required documentation for consideration for registration with the U.S. Environmental Protection Agency.

Propargyl bromide was one component of Tri-Zone, a previously available material, but the explosive nature of the material led to its removal from the commercial market. A commercial entity recently developed a new formulation that could be handled safely. The results of these trials demonstrate that propargyl bromide is as effective as methyl bromide at controlling plant-parasitic nematode populations at planting and after one growing season and could be a viable alternative to methyl bromide based on efficacy. Unfortunately, there is currently no known registrant, and work to commercialize this product appears to have stopped. Until there is interest in commercializing this material, it will not be available as an alternative to methyl bromide.

Sodium azide was previously available in a granular formulation and was shown to be an ineffective nematicide under field conditions (Kelley and Rodriguez-Kabana 1978). Recent research on annual crops has indicated that new liquid formulations might have more nematicidal activity (Gerik et al. 2002, Rodriguez-Kabana et al. 2004). Under vineyard replant conditions, sodium azide in the Agrizide formulation demonstrated efficacy not significantly different from methyl bromide in samples taken at planting. However, after the first growing season, root-knot nematode populations were significantly higher on susceptible varieties and rootstocks in the sodium azide plots than in the methyl bromide-treated plots. This finding suggests that although nematode populations were initially reduced to low levels, the residual root-knot nematode populations were sufficient to increase to significantly higher levels after one growing season. Since the nematode populations would be expected to continue to increase each growing season, sodium azide at the tested application rates does not appear to provide adequate nematode control for a perennial crop. Since sodium azide is a general biocide, it is likely that concentrations sufficient to kill nematodes in the old roots were not delivered to the deeper soil depths under the soil conditions present in these two trials. New formulations and new application protocols have recently been developed and tested in annual crops (Gerik et al. 2002, Rodriguez-Kabana et al. 2004). These formulations may have greater or lesser efficacy under vineyard replant conditions than observed for the formulation used in the replant trials reported here.

Alternative application technologies. In earlier trials of drip application of nematicides for bulb crops in fine-textured soils, nematode-control efficacy varied relative to the control achieved with the industry standard shank-injected protocols (Westerdahl et al. 1993, 2003). Delivery of nematicidal materials to soil depths greater than 60 cm is critical, even in annual crops, to achieve acceptable nematode control and improve crop growth (Westphal et al. 2004). In recent trials, drip application of fumigants in sandy loam soils has provided good results when used with both current commercially available compounds, such as 1,3-D and chloropicrin, and those not yet on the market, such as iodomethane and propargyl bromide (Ajwa and Trout 2004, Schneider et al. 2003). Improvements in application equipment and methods, increased knowledge and understanding of fumigant distribution, and the moderately coarse soil texture likely contributed to the improved efficacy and consistency of nematode control following drip fumigation in the later trials. Drip application offers advantages, such as a closed-application system that limits worker exposure to fumigants during the application process and the potential to use the water front to move materials more uniformly through the soil profile than might be possible with shank injection (Ajwa and Trout 2004, Ajwa et al. 2002b).

In bed-fumigated crops, such as strawberry, the drip lines used to deliver the fumigant are often near the soil surface (Ajwa and Trout 2004). The bed is then covered with a plastic tarp before the fumigation is initiated. In perennial crops where plastic mulching is not a standard part of the cultural production practices, the drip lines used for fumigation were buried 25 cm deep to reduce volatilization loss of the fumigant. A water or metam sodium cap used both before initiation and after completion of the fumigation was also used to reduce the potential for fumigant loss. Consequently, much of the 0 to 30 cm soil sample is collected from soil that is above the drip tube delivering the fumigant. In the studies reported in this paper, a water cap was used when the fumigants were expected to have sufficient volatility to move and treat the soil above the drip line. However, weed control in plots receiving water caps was not acceptable in either trial, whereas plots receiving a metam sodium cap showed good weed control. The water cap may have moved the chemicals deeper in the soil and reduced their efficacy to control weeds. Our previous research on fumigant distribution in soil found that application of a water cap (12 mm water) reduced fumigant concentration above the buried tape by 50 to 70% (data not shown). Nonetheless, our results found that concentrations of Midas and propargyl bromide were sufficient to control nematodes in the top 30 cm of soil without use of a metam sodium cap.

The sodium azide and InLine plots had slightly higher nematode populations in the first trial. Combining the InLine treatment with a metam sodium cap in the 2003 trial resulted in improved nematode control. Although shallow placement of Agrizide gave better weed control prior to planting than deep placement, nematode control at the time of planting and after the first growing season was better in plots treated with sodium azide delivered through standard-depth drip lines. A combination of sodium azide applied through the standard-depth drip lines coupled with an appropriate herbicide/nematicide cap treatment might provide better control of both nematodes and weeds.

The continued availability of metam sodium applied through microsprays is not certain because of the potential for off-gassing, which could trigger regulatory intervention. If a metam sodium cap was not available, then other means to seal the soil surface and treat the soil above the drip tape would be needed to meet the level of control provided by methyl bromide. One possibility would be to tarp the soil before the fumigation, as is done in bedded crops such as strawberry. That would impose an additional cost (about \$2000 ha^{-1} for purchase, placement, removal, and disposal of standard plastic mulch), since plastic mulch is not a standard part of the vineyard production system.

Resistance. Resistant rootstocks can be a good tool for managing vineyard nematode populations. Selection of an appropriate rootstock requires accurate identification of the nematode genera present and availability of correspondingly resistant rootstock. After one growing season, own-rooted Thompson Seedless supported higher populations of both root-knot and citrus nematodes in the untreated control plots than did Harmony, Freedom, or 1103P. Long-term studies have shown that, over time, citrus nematode populations can increase on these rootstocks while root-knot nematode populations remain low (Schneider et al. 2003). Identifying a rootstock that is resistant to the entire complex of nematodes and pathogens often found in old vineyards and provides the desired cultural characteristics for the chosen scion can be difficult or impossible (Anwar et al. 2002, McKenry et al. 2001a,b).

Integrated approaches. Combinations of management strategies, such as resistant rootstocks coupled with soil fumigation, should provide extra measures of nematode control. While the effect of the resistant rootstocks is clear in the untreated plots, the additional benefit was not observed in nematode populations after one growing season in the plots receiving chemical treatments. It is likely that a highly effective nematicide, such as 1,3-D, obscures the contribution of the resistant rootstocks. It is possible that these contributions will become more evident later in the life of the vineyard. Ongoing vineyard replant trials seek to further investigate the long-term advantages of combination treatments.

Plant growth. Nematode and weed control are not the only measures of a successful alternative to methyl bromide for vineyard replant. Vine growth reflects the plant's response to the management strategies. In the field trials reported here, vine growth is clearly not determined solely by nematode population densities. Although plants were much larger in the 2003 trial than in the 2001 trial, similar trends are observed for the own-rooted Thompson Seedless, Thompson Seedless on Freedom, and Autumn Royal on Harmony. Despite equivalent nematode control in plots treated with iodomethane, 1,3-D, chloropicrin, and methyl bromide, vine growth was significantly greater during the first growing season in plots treated with methyl bromide. Several treatments contained significant levels of chloropicrin, a compound with fungicidal properties that would be expected to control soilborne pathogens. In the trials described here, methyl bromide resulted in better plant growth than did combinations of nematicides, fungicides, and herbicides. Only propargyl bromide resulted in growth of own-rooted Thompson Seedless comparable to methyl bromide.

Yield. Differences in first year vine growth did not result in comparable differences in early fruit yield. In the first year of harvestable fruit, the impact of soil fumigation was evident on own-rooted Thompson Seedless. The contribution of the nematode resistant rootstock, Freedom, to the first year of fruiting, can be seen by comparing the yield of the own-rooted Thompson Seedless and Thompson Seedless on Freedom in the untreated and sodium azide-treated plots. Thompson Seedless on Freedom resulted in fruit yield comparable to methyl bromide even in the untreated plots. In the second year of fruit harvest, there was no difference across treatments for any rootstock. This one year should be viewed with both interest and caution. While these data show a lack of fumigant impact on yield for some variety/rootstock combinations in this field in some years, the information cannot be extrapolated to all variety/rootstock combinations in all fields for all years.

These results indicate the difficulty to find alternatives to methyl bromide for vineyard replant. The question remains as to what components of the vineyard ecosystem, biotic or abiotic, are positively impacted by methyl bromide, but not as significantly impacted by other fumigants. Further, these trials illustrate the importance of variety and rootstock differences in their responses to potential methyl bromide alternatives. Growth of own-rooted Thompson Seedless, Thompson Seedless on Freedom, and Autumn Royal on Harmony in the first year was reduced in untreated soil in both trials to less than 25% of the weight of vines grown in methyl bromide fumigated soil. Merlot on 1103P responded similarly in the 2003 trial but not in the 2001 trial. Yield response did not always reflect prior differences in vine growth. These trials illustrate that for these two vineyard replant situations, factors controlled by methyl bromide other than root-knot and citrus nematodes are differentially impacting vine growth.

Methyl bromide and 1,3-D have been the preferred control strategies for vineyard replant disorder for decades. Application technologies and crop production systems have likely been optimized around the use of these fumigants. Potential methyl bromide alternatives do not yet have this body of practical knowledge and experience.

Conclusions

New chemicals and alternative application technologies for existing chemicals provided good results as alternatives to methyl bromide for nematode control in vineyard replant situations. Iodomethane in combination with chloropicrin has demonstrated good efficacy for nematode control. Propargyl bromide, while effective for controlling nematodes, is not currently under commercial development. Sodium azide requires further research to evaluate different formulations and rates for vineyard replant.

Improved efficacy of drip application of fumigants is an important development in the search for methyl bromide alternatives. Combination of drip-applied fumigants with a supplemental treatment for the soil surface gave control of nematodes and weeds equal to methyl bromide in vineyard replant. Drip-applied 1,3-D + chloropicrin is commercially available for field and vegetable crops at label rates up to 243 L ha^{-1}. The higher rates used in this study, which provided good nematode control, are labeled only for pineapple at this time. The shank-injected formulation, Telone C-35, is labeled for fruit and nut crops at the rate used in this study. The ability to use either shank injection or drip application for some fumigants provides greater flexibility to select the best application method based on equipment availability and soil conditions. Use of 1,3-D is restricted in California by annual township caps, which might limit the availability of this alternative for vineyard replant situations, especially in townships with significant amounts of strawberry or vegetable use of 1,3-D.

Selection of appropriate resistant rootstocks requires knowledge of the pest, nematode, and pathogen complex present in a given vineyard and must be balanced with desired cultural characteristics such as vigor and drought tolerance. Vineyards often have a greater diversity of pest problems than rootstocks have resistance. Unless the diversity of pests and nematodes in the vineyard is quite narrow, resistance alone will not be an adequate alternative to methyl bromide.

First-year vine growth in these two vineyards is impacted by factors other than the plant-parasitic nematode populations. These factors appear to differentially impact the variety/rootstock combinations and to vary between the two sites. Reduced vine growth was not always correlated with reduced yield in the first and second years of fruit harvest.

Our trials indicate that 1,3-D + chloropicrin, iodomethane + chloropicrin, and propargyl bromide are good alternatives to methyl bromide for nematode control in sandy loam soils, but were not sufficient for management of all components of replant disorder at the rates used in this study, as reflected by reduced vine growth compared to methyl bromide-treated plots. Inability to positively determine the components contributing to vineyard replant disorder complicates the search for acceptable management strategies that can be used as alternatives to methyl bromide.

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