

Efficacy of 1,3-Dichloropropene Gelatin Capsule Formulation for the Control of Soilborne Pests

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The gelatin capsule (gel cap) formulation of 1,3-dichloropropene (1,3-D) is a new concept to reduce the environmental release, transport, and hazard potential of the use of 1,3-D to control soilborne diseases and nematodes. The objective of this study is to evaluate the biological efficacy of the 1,3-D gel cap formulation under laboratory and greenhouse trial conditions. Greenhouse experiments were carried out in suburbs of Beijing and Hebei Province of China in 2007 and 2008, focused mainly on tomato and *Bellis perennis* L. (daisy) crops. Results showed that 1,3-D gel cap application at a rate of 16.8 g of active ingredient m⁻² was as effective as 1,3-D liquid injection treatment. Crop yields in plots treated with 1,3-D gel cap and 1,3-D liquid were significantly higher than those in untreated control. The present study confirms that the 1,3-D gel cap formulation is a promising new formulation with good field efficacy.

KEYWORDS: 1,3-Dichloropropene; gelatin capsule formulation; soilborne pests; control effect

INTRODUCTION

The use of methyl bromide (MeBr) as a soil fumigant to control soilborne pests is being phased out internationally because it is a stratospheric ozone-depleting compound. Effective alternatives to MeBr must be developed. A potential alternative to MeBr is 1,3-dichloropropene (1,3-D). 1,3-D is an effective nematicide with some fungicidal activity (1, 2). It is commonly applied alone as a fumigant to control nematodes or combined with chloropicrin to control soilborne diseases and weeds (3, 4). Because of large emission loss and off-site air pollution (5), agricultural use of 1,3-D in California and other regions of the world is currently limited to very low rates only under restricted conditions (6,7). In Florida and in Prince Edward Island, Canada, 1,3-D is restricted because of reported seepage into groundwater (8). Thus, effective and environmentally friendly application methods are needed to reduce the environmental and human risk potential of 1,3-D and to establish 1,3-D as an effective alternative fumigant to MeBr for agricultural crops.

Many experiments have been conducted to evaluate alternative methods to reduce the environmental release of 1,3-D. Schneider et al. reported that 1,3-D emission could be reduced by increasing application depth (9), and Wang et al. reported that the use of subsurface drip irrigation with a lower dosage rate of 1,3-D also reduced emission potentials (7). The use of agricultural films (i.e., tarps or mulch) had also been studied as a mitigation strategy for reducing 1,3-D emissions. Field data showed that virtually impermeable film (VIF) is more effective in reducing 1,3-D emissions than high-density polyethylene (HDPE) tarp (10). However, the use of agricultural films is also limited due to higher

cost and disposal difficulties. 1,3-D emissions can be reduced by applying composted animal manure to fields (11). Zheng et al. used thiourea to construct a reactive surface barrier (RSB) on the soil surface to reduce 1,3-D volatilization (12). Gan et al. reported that surface application of thiosulfate fertilizers may also be a feasible and effective strategy to minimize 1,3-D emission (13). Furthermore, the application of a surface water seal has been shown to reduce 1,3-D emissions by forming a high water content layer at the soil surface as a diffusion barrier (14–19).

New formulations of 1,3-D allow the use of different application methods that are more effective, less costly, and more environmentally friendly (8). 1,3-D is usually applied by injection or by drip application (4, 20). The gelatin capsule (gel cap) formulation of 1,3-D is a new concept to reduce 1,3-D emissions and leaching and reduces worker and bystander exposure. 1,3-D gel cap can be stored for a long time without breakdown and is easier to use in the soil without the use of special equipment. 1,3-D is a strong skin irritant and has a potential inhalation hazard, requiring personal protective equipment when applied in liquid form. This is a limitation to its adoption in China because most farmers apply the fumigants by themselves. The gel cap formulation offers a good solution to these application constraints.

The objective of the study was to determine the efficacy of the 1,3-D gel cap formulation to soilborne diseases and nematodes by means of both laboratory bioassay and greenhouse trials. The greenhouses used by farmers in northern China are usually constructed in the field by excavating the soil to approximately 50 cm, forming a wall about 2 m tall on one side, and then covering the excavated area with plastic films over the wall supported by a frame system and at the soil level on the opposite side of the wall. The area of a greenhouse was about 700 m².

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MATERIALS AND METHODS

1,3-D Gelatin Capsule. 1,3-D gel caps were produced on a common capsule machine modified by adding sealing equipment to eliminate 1,3-D emission during processing. Technical grade 1,3-D (purity = 95%) was provided by Beijing Zhongzhikehua Agricultural Technology Co. Ltd. The outer gelatin shell (0.8 mm thickness) of the gel cap was supplied by Qinhai Gelatin Co. Ltd. The average volume of a gel cap is 1 mL, and the average weight of 1,3-D in a gel cap is 0.8 g.

Bioassays in the Soil Boxes. Soil samples from the field experiment were collected from the top 15 cm in the greenhouse at Langfang, Hebei Province, which were heavily infested with the root-knot nematodes and pathogens. The soil characteristics were as follows: pH 7.6; soil classification, 83.4% sand, 15.1% silt, 1.6% clay, and 1.5% organic materials.

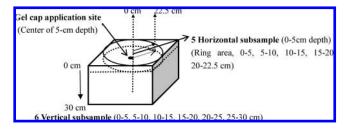


Figure 1. Illustration of box bioassay.

 Table 1.
 Treatments in Field Trials

year and crop	fumigants and formulation	l active ingredient	dose (g of ai m ⁻²	abbreviation of) treatment
trial 1 summer 2008	1,3-D gel cap 1,3-D liquid	1,3-D 1,3-D	16.5 16.5	1,3-D gel cap 16.5 1,3-D liquid 16.5
tomato	untreated control			СК
trial 2	1,3-D gel cap	1,3-D	8.4	1,3-D gel cap 8.4
fall 2007			16.8	1,3-D gel cap 16.8
Bellis	1,3-D liquid	1,3-D	8.4	1,3-D liquid 8.4
perennis l	L.			
			16.8	1,3-D liquid 16.8
	MeBr liquid	MeBr	50	MB 50
	untreated control			СК



Figure 2. Example of the gel cap application.

Table 2.	Crop and	Growth	Calendar	for the	Vegetables
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For the fumigation treatment experiment, a single 1,3-D gel cap was placed into a box ($45 \times 40 \times 30$ cm) containing the soil mentioned above [1.14 g cm⁻³ and 20.5% (w/w) moisture]. The gel cap was placed in soil 5 cm under the top in boxes covered with the PE films, where the fumigation period lasted for 15 days at 20–34 °C. Untreated control (CK, a box containing the same soil without a 1,3-D gel cap) was placed in the same conditions with the treated boxes. Experiments were conducted with four replicates.

After the fumigation treatments, soil samples were collected vertically at the sites from the top center to 5, 5-10, 10-15, 15-20, 20-25, and 25-30 cm soil boxes, respectively. Similarly, additional samples were collected horizontally within the soil layer (0-5 cm in depth), where there are ring sampling areas 5, 5-10, 10-15, 15-20, and 20-22.5 cm from the box center to the edge. Examples of the soil collection positions are illustrated in **Figure 1**.

The nematodes and fungal pathogens in the sampled soils were separated for evaluating the fumigation efficacy of the 1,3-D gel cap. Nematodes were separated from 100 g soil subsamples by centrifugation. The semiselective solid medium described by Masago was used for *Phytophthora* and *Pythium* detection: selective inhibition of *Pythium* spp. on medium for direct isolation of *Phytophthora* spp. from soils (21). A semiselective medium described by Komada was used for *Fusarium* detection: development of selective medium for quantitative isolation of *Fusarium oxysporum* from natural soil (22).

Field Trial. In 2007 and 2008, two experiments were conducted in greenhouses located in Hebei Province (Buying Village, Langfang City; soil pH 7.6; soil organic matter content of 1.5%; 83% sand, 15% silt, and 1.6% clay) and from a location within a Beijing suburb (Tujing Village, Malianwa District; soil pH 8.3; soil organic matter of 2.8%; 59% sand, 37% silt, and 3.9% clay). Both test sites are situated in areas of intensive vegetable production and MeBr consumption in China. As a result of consecutive cultivation and less effective rotation, the occurrence of nematodes and soilborne diseases has become more severe in these regions. The test crops were tomato and *Bellis perennis* L. (daisy) at the Beijing and Hebei sites, respectively.

A summary of the field trial is shown in Table 1. Both 1,3-D liquid and gel cap were applied at a dosage of 16.5 g of active ingredient (ai) m^{-2} in tomato fields, respectively, whereas dosages of 8.4 and 16.8 g of ai m⁻² were used in the *B. perennis* L. fields. MeBr was applied at a dosage of 50 g of ai m^{-2} in *B. perennis* L. fields. The gel caps were applied to field soil at a 10 cm depth by forming a hole and inserting a gel cap without special application tools, similar to planting a seed. An example of the gel cap application is presented in Figure 2. 1,3-D liquid was injected to the soil at the 10 cm depth via a manual injection machine (model JM-C), which was obtained from Dalian Jinmei Soil Disinfection Equipment Development Co. Ltd. The manual injection machine uses the working principle of the piston. Under the power of the piston, the fumigant in reservoir barrels was injected into soil through the piston cylinder and vent valve after the bar had been manually pressed. MeBr was applied at the soil surface through a plastic tube with many holes on its surface. Each plot area was designed to 24 m² and arranged using a randomized block design. Each treatment was repeated four times, and the treated soil areas were covered with 0.08 mm polyethylene plastic film.

Soil treatments were carried out on October 9, 2007, in Hebei and on July 25, 2008, in Beijing, respectively. Fumigation areas were undisturbed for 15 days in Hebei and for 10 days in Beijing. To achieve a complete diagnosis of plant diseases that could occur in the experimental crops, the nematodes pathogen (*Fusarium* spp. and *Phytophthora* spp.) population densities of each plot were evaluated before and after treatment, and the final evaluation was carried out at harvest time. Soil samples were collected at soil depths of 5, 10, 15, and 20 cm from each plot separately. A subsample (100 g) was used for nematode analysis by centrifugation, and a 5 g subsample was used for soil pathogen analysis. *Fusarium* and

site	crop	seedling	transplanting	beginning of harvesting	finish of the season	date of treatment	sampling after treatment	sampling at harvest
Beijing	tomato	July 4, 2008	Aug 15, 2008	Oct 5, 2008	Dec 5, 2008	July 25, 2008	Aug 3, 2008	Dec 5, 12
Hebei	<i>Bellis perennis</i> L.		Nov 3, 2007	Dec 3, 2007	April 7, 2008	Oct 9, 2007	Oct 24, 2007	April 7, 2008

Phytophthora spp. were collected according to Komada's method (21) and Masago's method (22), respectively.

Cultivation information is listed in **Table 2**; traditional cultivation techniques were adopted in all plots. Marketable yields of tomato and *B. perennis* L. were recorded separately.

Data Analysis. Control effect (CE) after fumigation on pathogens is expressed as eq I

control effect = CE =
$$\left(1 - \frac{P_{\rm T}}{P_{\rm CK}}\right) \times 100$$
 (I)

where $P_{\rm CK}$ is the population density of pathogen in untreated control and $P_{\rm T}$ is the population density of pathogen in the fumigation treatment.

CE after fumigation on nematodes is expressed as eq II

control effect = CE =
$$\left(1 - \frac{S_{\rm T}}{S_{\rm CK}}\right) \times 100$$
 (II

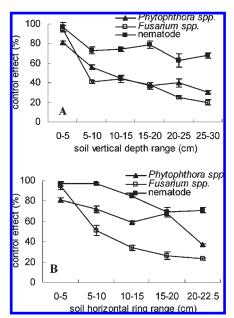


Figure 3. Efficacy of 1,3-D gel cap on soilborne pests in soil box: (A) vertical depth; (B) horizontal ring range.

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where S_{CK} is the number of surviving nematodes in untreated control and S_{T} is the number of surviving nematodes in the fumigation treatment.

The data were statistically analyzed according to Duncan's multiplerange test with the Statistical Analysis System (SAS) computer program.

RESULTS AND DISCUSSION

Results of Bioassay Using the Soil Box. The bioassay results of the 1,3-D gel cap are shown in Figure 3. The highest control effects (97% for nematode, 95% for Phytophthora spp., and 81% for Fusarium spp.) of 1.3-D on nematodes and pathogens occurred near the application site (0-5 cm depth). However, the control effects decreased with the vertical depth and horizontal width away from the site of treatment. The lowest observable control effect on nematodes was still above 63%. These results indicate that the 1,3-D gel cap formulation has good nematode control. The control effects on Fusarium spp. were below 51% in all soil samples except at the application site. The control effects on Phytophthora spp. were similar to those on Fusarium spp. in vertical samples; however, the control effects pn Phytophthora spp. were better than those on Fusarium spp. in horizontal samples. The results indicated that the 1,3-D gel cap also has efficacy to control soil pathogens, but the control effect was lower than that shown for the control of nematodes.

Tomato Field Trial Results in Beijing (2008). The efficacy of 1,3-D on the treatment of nematodes in the tomato field is shown in **Table 3**. The control effects of the 1,3-D gel cap on nematodes were above 80%, and there is no significant difference between the gel cap and the injection method. During the period of harvest time, nematodes in soil populations had increased, but the population densities in gel cap and injection treatments were significantly lower than that in the untreated control (CK) at that time. The results confirmed that the 1,3-D gel cap gave satisfactory control of nematodes comparable to the soil injection treatment method.

After treatment, the soil populations of *Fusarium* and *Phy-tophthora* spp. were all significantly reduced (>75%) by the 1,3-D gel cap and direct injection treatments compared with the CK (**Tables 4** and **5**). At harvest, the population of *Fusarium* spp. was still significantly reduced by two fumigation treatments as compared with the CK (**Table 4**); however, the populations of

Table 3. Effect of Soil Fumigation on Nematode Population in Tomato Field (Number of Nematodes per 100 g of Soil)

			Sample 2 h before	e Treatment (July 25	, 2008)			
treatment	5 cm		10 cm		15 cm		20 cm	
1,3-D gel cap 16.5	117		225		208		183	
1,3-D liquid 16.5 CK	208 242		217 642		350 292		158 492	
		:	Sample 9 Days aft	er Treatment (Aug 3	8, 2008)			
treatment	no.	CE (%)	no.	CE (%)	no.	CE (%)	no.	CE (%)
1,3-D gel cap 16.5	17 b ^a	85	0 b	100	0 b	100	0 b	100
1,3-D liquid 16.5	17 b	88	0 b	100	8 b	92	25 b	81
CK	133 a		558 a		383 a		467 a	
			Sample at Harves	st, 134 Days (Dec 5,	2008)			
treatment	no.	CE (%)	no.	CE (%)	no.	CE (%)	no.	CE (%)
1,3-D gel cap 16.5	58 b	88	108 c	83	75 b	83	83 b	85
1,3-D liquid 16.5	83 b	83	300 b	42	100 b	81	100 b	83
CK	475 a		575 a		550 a		558 a	

^a Treatments with the same letter were not significantly different according to Duncan's multiple-range test at the 5% significance level.

Table 4. Effect of Soil Fumigation on Fusarium Specie	es in Tomato Field [Colony-Forming Units (CFU) per Gram of Soil]
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			Sample 2 h befor	e Treatment (July 25	5, 2008)			
treatment	5 cm 10 cm				15 cm		20 cm 516 302 442	
1,3-D gel cap 16.5 1,3-D liquid 16.5 CK	318	258 151 318 382 518 769			198 309 162			
			Sample 9 Days af	ter Treatment (Aug	3, 2008)			
treatment	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)
1,3-D gel cap 16.5 1,3-D liquid 16.5 CK	9 b ^a 0 b 338 a	89 100	0 b 4 b 320 a	100 99	20 b 22 b 133 a	86 78	9b 71b 311 a	97 73
			Sample at Harve	st, 134 Days (Dec 5	, 2008)			
treatment	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)
1,3-D gel cap 16.5 1,3-D liquid 16.5 CK	97 c 309 b 550 a	70 27	13 b 0 b 150 a	88 95	0 b 43 b 287 a	99 81	44 b 7 b 587 a	92 96

^a Treatments with the same letter were not significantly different according to Duncan's multiple-range test at the 5% significance level.

Table 5. Effect of	of Soil Fumgiation on	Phytophthora Species in	Tomato Field [Color	ny-Forming Units	(CFU) per Gram of Soil]

			Sample 2 h before	e Treatment (July 25	5, 2008)			
treatment	5 cr	n	10 ci	m	15 cm	l	20 cm	
1,3-D gel cap 16.5 1,3-D liquid 16.5 CK	410 286 508	7	3938 2904 5362		3844 2740 4211		3129 2480 3011	
			Sample 9 Days aft	er Treatment (Aug	3, 2008)			
treatment	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)
1,3-D gel cap 16.5 1,3-D liquid 16.5 CK	647 b ^a 430 b 5520 a	88 92	670 b 627 b 3010 a	78 79	590 b 490 b 4380 a	86 88	1007 b 583 b 3400 a	70 83
			Sample at Harves	st, 134 Days (Dec 5	, 2008)			
treatment	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)
1,3-D gel cap 16.5 1,3-D liquid 16.5 CK	1816 a 1062 b 2578 a	18 51	2853 a 1284 a 1989 a	12 37	2138 a 1969 a 2009 a	14 15	1758 a 1687 a 1931 a	5 14

^a Treatments with the same letter were not significantly different according to Duncan's multiple-range test at the 5% significance level.

Phytophthora spp. were not significantly different in all treatments (Table 5).

Table 6. Effects of Soil Fumigation on Root-Knot Index and Yield of Tomato, 2008

Nematode gall index was significantly reduced by both 1,3-D treatments at final harvest (Table 6). Tomato yields treated with the 1,3-D gel cap and 1,3-D injection were significantly higher than that in the CK (Table 6). There was no significant difference in tomato yields between 1,3-D gel cap treatment and 1,3-D injection at the Beijing site (Table 6). The results suggested that the tomato yield was significantly increased after fumigation with 1,3-D.

Results of B. perennis L. Field Trial in Hebei (2007). The effect of soil disinfestation on nematodes in the B. perennis L. field is shown in Table 7. After fumigation, populations of nematode were all reduced from prefumigation levels and were significantly less for all fumigated treatments as compared with the CK;

treatment marketable yield (ton ha^{-1}) gall index 1,3-D gel cap 16.5 56 a^a 18 b^b 1,3-D liquid 16.5 57 a 10 b CK 50 b 55 a

^a Treatments with the same letter were not significantly different according to Duncan's multiple-range test at the 5% significance level. ^b The galling index in tomato was divided into five grades: 0 for none and 4 for above 75% with root-knot.

control effects for all fumigated treatments were all >90%. By final harvest of B. perennis L., nematode soil populations in CK were significantly less than prefumigation levels; the lower

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Table 7. Effect of Soil Fumigation on	Nematode Population in Bellis p	perennis L. Field (Number	of Nematodes per 100 g of Soil)

			Sample 2 h befor	re Treatment (Oct 9,	2007)			
treatment	5 c	m	10 c	m	15 cm		20 cm	
1,3-D gel cap 8.4	233	33	339	9	1900		1208	
1,3-D gel cap 16.8	17	1	211	0	1572		1065	
1,3-D liquid 8.4	313	35	286	1	1434		889	
1,3-D liquid 16.8	200	06	120	0	588		649	
MB 50	129	95	94	5	820		640	
СК	25	0	161	5	1271		1165	
		ę	Sample 15 Days af	ter Treatment (Oct 2	24, 2007)			
treatment	no.	CE (%)	no.	CE (%)	no.	CE (%)	no.	CE (%)
1,3-D gel cap 8.4	177 b ^a	95	172 b	92	38 b	97	40 b	96
1,3-D gel cap 16.8	7 b	100	7 b	100	13 b	99	0 b	100
1,3-D liquid 8.4	7 b	100	7 b	100	9 b	99	7 b	99
1,3-D liquid 16.8	0 b	100	0 b	100	0 b	100	0 b	100
MB 50	8 b	100	10 b	99	8 b	99	9 b	99
СК	2893 a		1956 a		1358 a		1065 a	
			Sample at Harves	st, 180 Days (April 7	, 2008)			
treatment	no.	CE (%)	no.	CE (%)	no.	CE (%)	no.	CE (%)
1,3-D gel cap 8.4	183 a	54	50 b	80	83 a	57	17 b	83
1,3-D gel cap 16.8	42 b	90	42 b	83	33 b	83	25 b	75
1,3-D liquid 8.4	283 a	29	67 b	73	58 b	70	58 a	42
1,3-D liquid 16.8	8 b	98	17 b	93	17 b	91	8 b	92
MB 50	86 b	79	63 ab	75	48 b	75	68 a	32
CK	400 a		250 a		192 a		100 a	

Table 8. Effect of Soil Fumigation on Fusarium Species in Bellis perennis L. Field (Colony-Forming Units per Gram of Soil)

			Sample 2 h before	e Treatment (Oct 9,	2007)			
treatment	5 cm	1	10 cr	n	15 cm		20 cm	
1,3-D gel cap 8.4	593	3	270	0	580		233	
1,3-D gel cap 16.8	763	}	613	3	2020		2140	
1,3-D liquid 8.4	1093	}	138	7	1307		977	
1,3-D liquid 16.8	610)	507	7	377		180	
MB 50	167		193		130		200	
СК	400)	100	7	303		103	
		Si	ample 15 Days aft	er Treatment (Oct 2	24, 2007)			
treatment	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)
1,3-D gel cap 8.4	116 b ^a	86	104 b	82	167 b	68	367 b	68
1,3-D gel cap 16.8	93 b	86	76 b	88	51 b	90	71 bc	89
1,3-D liquid 8.4	102 b	87	113 b	82	100 b	81	213 b	79
1,3-D liquid 16.8	133 b	89	111 b	84	87 b	84	120 b	93
MB 50	16 c	98	9 c	99	11 c	98	22 c	96
СК	1082 a		667 a		533 a		1089 a	
			Sample at Harves	t, 180 Days (April 7	, 2008)			
treatment	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)
1,3-D gel cap 8.4	1289 a	0.00	667 a	29	696 a	0	649 a	0
1,3-D gel cap 16.8	818 a	0.00	362 a	62	289 a	50	129 a	50
1,3-D liquid 8.4	389 ab	0.00	353 a	62	391 a	32	413 a	0
1,3-D liquid 16.8	60 c	73	9 b	99	16 b	97	42 b	84
MB 50	2 d	99	0 b	100	0 b	100	7 b	97
CK	224 b		942 a		576 a		256 a	

^a Treatments with the same letter were not significantly different according to Duncan's multiple-range test at the 5% significance level.

 Table 9. Effect of Soil Fumigation on Phytophthora Species in Bellis perennis L. Field [Colony-Forming Units (CFU) per Gram of Soil]

			Sample 2 h before	e Treatment (Oct 9,	2007)			
treatment	5 cm	ı	10 cn	n	15 cm		20 cm	
1,3-D gel cap 8.4	2647	7	2193		1597		947	
1,3-D gel cap 16.8	2840)	2193	}	2613		4000	
1,3-D liquid 8.4	1633	3	2013	}	1840		2670	
1,3-D liquid 16.8	2660)	2540)	2380		1733	
MB 50	2627	7	1393	}	1560		1853	
СК	1013	3	1133	}	1587		1920	
		S	ample 15 Days afte	er Treatment (Oct 2	4, 2007)			
treatment	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)
1,3-D gel cap 8.4	1533 b ^a	45	1307 b	51	1044 b	61	1127 b	52
1,3-D gel cap 16.8	1524 b	45	1533 b	48	1169 b	62	1062 b	62
1,3-D liquid 8.4	1369 b	49	1000 b	61	1176 b	53	1053 b	60
1,3-D liquid 16.8	1044 b	59	1027 b	59	751 b	73	811 b	66
MB 50	13 c	99	11 c	100	7 c	100	18 c	99
СК	2796 a		2636 a		2693 a		2680 a	
			Sample at Harvest	, 180 Days (April 7	, 2008)			
treatment	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)	CFU	CE (%)
1,3-D gel cap 8.4	2391 a	6	2391 a	0	2169 a	0	1840 a	0
1,3-D gel cap 16.8	2202 a	14	2400 a	0	1296 a	23	1144 a	27
1,3-D liquid 8.4	1964 a	23	1662 a	0	1424 a	16	1142 a	27
1,3-D liquid 16.8	1153 a	55	880 a	36	1407 a	17	678 a	56
MB 50	9 b	100	9 b	99	7 b	100	40 b	97
CK	2556 a		1382 a		1689 a		1558 a	

^a Treatments with the same letter were not significantly different according to Duncan's multiple-range test at the 5% significance level.

survival rates of nematode may be due to irrigation when sampling. Higher soil-water content affected nematode egg hatching and second-instar survival (23), but populations of nematode were still significantly less following 1,3-D gel cap and injection treatment at a dosage of 16.8 g of ai m^{-2} compared with the CK, and control effects for the two treatments were all >75%.

The effect of soil disinfestation on *Fusarium* spp. in the *B. perennis* L. field is shown in **Table 8**. After disinfestations, the quantity of *Fusarium* spp. was significantly reduced by all of the fumigant applications. Methyl bromide more effectively reduced *Fusarium* spp. population than 1,3-D. At final harvest, *Fusarium* spp. populations in 1,3-D gel cap at two dosages and 1,3-D injection at 8.4 g of ai m^{-2} treatments were not significantly different from the CK treatment. The efficacy of MeBr to control *Fusarium* spp. was found to be nearly 100%. These results indicate that the 1,3-D gel cap at high dosage has the capacity to control *Fusarium* spp., but the efficacy was still less than that of MeBr.

The effect of soil fumigation on *Phytophthora* spp. in the *B. perennis* L. field is shown in **Table 9**. After disinfestations, populations of *Phytophthora* spp. were reduced by all of the fumigant applications in the trial, but control effects of all 1,3-D treatments were only about 50%. At final harvest period, *Phytophthora* spp. populations in all 1,3-D treatments were not significantly different from the CK treatment, but control effects of MeBr on nematode were nearly 100% at harvest time. The results indicated that 1,3-D has low activity to control *Phytophthora* spp.

B. perennis L. yields in plots treated with 1,3-D gel cap at rates of 8.4 and 16.8 g of ai m^{-2} and with 1,3-D injection at a rate of 16.8 g of ai m^{-2} were significantly greater than that in the CK (**Table 10**) and showed no significant differences compared with MeBr treatment. There was no yield difference between plots treated with 1,3-D injection at a rate of 8.4 g of ai m^{-2} and CK.

Table 10. Effects of Soil Fumigation on the Yield of Bellis perennis L.

treatment	marketable yield (ton ha^{-1})
1,3-D gel cap 8.4	49.3 ab ^a
1,3-D gel cap 16.8	51.4 ab
1,3-D liquid 8.4	47.2 bc
1,3-D liquid 16.8	50.5 ab
MB 50	52.8 a
СК	45.0 c

^aTreatments with the same letter were not significantly different according to Duncan's multiple-range test at 5% the significance level.

The data from laboratory bioassay and field trials indicated that 1,3-D gel cap can effectively control nematodes and is partially effective for pathogens, similar to 1,3-D injection application. The results showed that the gel caps are a promising new formulation and that additional study is needed to determine their performance with respect to emissions and leaching.

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LITERATURE CITED

- Duniway, J. M. Status of chemical alternatives to methyl bromide for pre-plant fumigation of soil. J. Phytopathol. 2002, 92, 1337–1343.
- (2) Martin, F. N. Development of alternative strategies for management of soilborne pathogens currently controlled with methyl bromide. *Annu. Rev. Phytopathol.* 2003, *41*, 325–350.
- (3) Hutchinson, C. M.; McGiffen, M. E.; Sims, J. J.; Becker, J. O. Fumigant combinations for *Cyperus esculentum* L control. *Pest Manag. Sci.* 2003, 60, 369–374.
- (4) Minuto, A.; Gullino, M. L.; Lamberti, F.; D'Addabbo, T.; Tescari, E.; Ajwa, H.; Garibaldi, A Application of an emulsifiable mixture of 1,3-dichloropropene and chloropicrin against root knot nematodes

and soilborne fungi for greenhouse tomatoes in Italy. *Crop Prot.* 2006, *25*, 1244–1252.

- (5) Van den Berg, F.; Roos, A. H.; Tuinstra, L. G. M. Th.; Leistra, M. Measured and computed concentrations of 1,3-dichloropropene and methyl isothiocyanate in air in a region with intensive use of soil fumigants. *Water, Air Soil Pollut.* **1994**, *78*, 247–264.
- (6) California Department of Food and Agriculture. Use of Pesticide Suspended by CDFA; news release 90-45; CDFA: Sacramento, CA, 1990.
- (7) Wang, D.; Yates, S. R.; Ernst, F. F.; Knuteson, J. A.; Brown, G. E. Volatilization of 1,3-dichloropropene under different application methods. *Water, Air Soil Pollut.* 2001, *127*, 109–123.
- (8) UNEP. 2006 Report of the Methyl Bromide Technical Options Committee, 2006 Assessment; United Nations Environment Program: New York, 2006; pp 108–109,ISBN: 978-92-807-2827-9.
- (9) Schneider, R. C.; Green, R. E.; Oda, C. H.; Sipes, B. S.; Schmitt, D. P. Reducing 1,3-dichloropropene air emissions in Hawaii pineapple with modified application methods. *Fumigants: Environmental Fate, Exposure, and Analysis*; ACS Symposium Series 652; American Chemical Society: Washington, DC, 1997; pp 95–103.
- (10) Thomas, J. E.; Allen, L. H.; McCormack, L. A.; Vu, J. C.; Dickson, D. W.; Ou, L. T. Diffusion and emission of 1,3-dichloropropene in Florida sandy soil in microplots affected by soil moisture, organic matter, and plastic film. *Pest Manag. Sci.* 2004, 60, 390–398.
- (11) Dungan, R. S.; Papiernik, S.; Yates, S. R. Use of composted animal manures to reduce 1,3-dichloropropene emissions. *J Environ. Sci. Health B* 2005, 40, 355–62.
- (12) Zheng, W.; Yates, S. R.; Papiernik, S. K.; Wang, Q. Reducing 1,3dichloropropene emissions from soil columns amended with thiourea. *Environ. Sci. Technol.* 2006, 40, 2402–2407.
- (13) Gan, J.; Becker, J. O.; Ernst, F. F.; Hutchinson, C.; Knuteson, J. A.; Yates, S. R. Surface application of ammonium thiosulfate fertilizer to reduce volatilization of 1,3-dichloropropene from soil. *Pest Manag. Sci.* 2000, *5*, 264–270.
- (14) Gao, S.; Trout, T. J. Using surface water application to reduce 1,3dichloropropene emission from soil fumigation. J. Environ. Qual. 2006, 35, 1040–1048.

- (15) Gao, S.; Trout, T. J. Surface seals reduce 1,3-dichloropropene and chloropicrin emissions in field tests. J. Environ. Qual. 2007, 36, 110– 119.
- (16) Gao, S.; Trout, T. J.; Schneider, S. Evaluation of fumigation and surface seal methods on fumigant emissions in an orchard replant field. J. Environ. Qual. 2008, 37, 369–377.
- (17) Ashworth, D. J.; Yates, S. R. Surface irrigation reduces the emission of volatile 1,3-dichloropropene from agricultural soils. *Environ. Sci. Technol.* 2007, 41, 2231–2236.
- (18) Yates, S. R.; Knuteson, J.; Ernst, F. F.; Zheng, W.; Wang, Q. Effect of sequential surface irrigations on field-scale emissions of 1,3dichloropropene. *Environ. Sci. Technol.* **2008**, *42*, 8753–8758.
- (19) Gao, S. D.; Qin, R. J.; McDonald, J. A.; Hanson, B. D.; Trout, T. J. Field tests of surface seals and soil treatments to reduce fumigant emissions from shank injection of Telone C35. *Sci. Total Environ.* 2008, 405, 206–214.
- (20) Gullino, M. L.; Camponogara, A.; Gasparrini, G.; Rizzo, V.; Clini, C.; Garibaldi, A. Replacing methyl bromide for soil disinfestation: the Italian experience and the implications for other countries. *Plant Dis.* 2003, 87, 1012–1021.
- (21) Komada, H. Development of a selective medium for quantitative isolation of *Fusarium oxysporum* from natural soil. *Rev. Plant Prot. Res.* 1975, 8, 114–125.
- (22) Masago, H.; Yoshikawa, M.; Fukada, M.; Nakanishi, N. Selective inhibition of *Pythium* spp. on a medium for direct isolation of *Phytophthora* spp. from soils and plants. *Phytopathology* **1977**, 67, 425–428.
- (23) Chen, L.; Wei, F.; Duan, Y.; Bai, C.; Huo, J.; Zhu, X. Effects of temperature and moisture on egg hatching and the second instars of *Meloidogyne incognita*. *Chinese Plant Prot.* 2009, 35 (2), 48–52.

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