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Enhanced Degradation of the Volatile Fumigant-Nematicides 1,3-D and Methyl Bromide in Soil[1]

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Abstract: The use of the gaseous fumigant-nematicide methyl bromide in agriculture is scheduled to be phased out in the year 2001. 1,3-Dichloropropene (1,3-D) in combination with chloropicrin and an herbicide is considered to be a viable alternative to methyl bromide for some crops. 1,3-Dichloropropene consists of two isomers, cis- and trans-1,3-D. A number of soil bacteria have been shown to initially degrade 1,3-D or one of its isomers, cis-1,3-D, via hydrolysis. Until recently, the degradation of cis- and trans-1,3-D in soils was considered to exhibit similar kinetics, with their degradation rates increasing with increases in soil temperature. Enhanced degradation of 1,3-D in soil from a site in Florida with a history of repeated annual applications of 1,3-D was observed in 1994. Biological hydrolysis was involved in the initial degradation of cis- and trans-1,3-D. The two isomers were degraded at different rates, with the trans isomer being degraded more rapidly than the cis isomer. Cis- and trans-1,3-D in soil from the control site were degraded at a similar rate but more slowly than in the enhanced soil. Methyl bromide in soils can be degraded through chemical hydrolysis and methylation to soil organic matter. Some methanotrophic bacteria and ammonia-oxidation bacteria during the oxidation of their primary substrates (methane and ammonia) also have the capacity to cooxidize methyl bromide to formaldehyde and bromide ion. It was recently observed that degradation of methyl bromide was stimulated in methanotrophic soils and in soils treated with ammonium sulfate. Soil methanotrophic bacteria and soil nitrifiers are apparently responsible for cooxidation of methyl bromide in methanotrophic and ammonia treated soils, respectively.

Key words: cis-1,3-D, degradation, enhanced soil, fumigant, methyl bromide, nematicide, nematode, nonfumigant, 1,3-D, 1,3-dichloropropene, Telone II, trans-1,3-D.

Methyl bromide (bromomethane, MeBr) and 1,3-dichloropropene (1,3-D) are used in agriculture as soil fumigants (DowElanco, 1996; Noling and Becker, 1994; Ou et al., 1995). Methyl bromide also is used as a space fumigant for commodities, structural pest control, and quarantine treatment and other regulatory purposes. The fate of the two chemicals for future use in agriculture may be in contrast to each other. Use of methyl bromide is scheduled to be phased out in the United States by 1 January 2001 (Noling and Becker, 1994), whereas 1,3-D is considered to be a viable alternative to MeBr for some crops (Anonymous, 1995; Stephens, 1996). Methyl bromide is a highly potent depleter of the stratospheric ozone layer (Watson, 1992); the disruption is due to the release of bromine atoms from MeBr into the stratosphere.

Both MeBr and 1,3-D are volatile, short-chained halogenated hydrocarbons. At ≥4 °C, MeBr is a gas (Hornsby et al., 1995), whereas 1,3-D at ambient temperature is a liquid (DowElanco, 1996; Hornsby et al., 1995). Both are fairly water-soluble (DowElanco, 1996; Hornsby et al., 1995; Yang, 1986) and subject to chemical hydrolysis in aqueous media (Gentile et al., 1989; McCall, 1987). 1,3-Dichloropropene consists of two isomers, the cis and trans forms. The two isomers have similar but not identical physical and chemical properties, with the cis isomer being slightly more volatile than the trans isomer, but less water-soluble (DowElanco, 1996; Yang, 1986). Telone II and Telone C17 (DowElanco, Indianapolis, IN) are the commercial formulations of 1,3-D. Telone II consists of 94% 1,3-D, and Telone C17 consists of 78% 1,3-D and 17% chloropicrin (DowElanco, 1996). 1,3-Dichloropropene does not have the multipurpose effectiveness of MeBr. It provides good control of nematodes but lacks herbicidal activity. The addition of chloropicrin (17%) to the formulation provides control of some soilborne fungal pathogens (Stephens, 1996). Telone C17 does not have any herbicidal properties; thus, an application of an

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herbicide is needed for Telone C17 to be an alternative to MeBr.

It has been known for more than 15 years that repeated field applications of some pesticides cause their enhanced degradation, and enhanced degradation may reduce pesticidal efficacy, resulting in crop failure (Racke and Coats, 1990). Two carbamate pesticides, EPTC (Rahman et al., 1979) and carbofuran (Felsot et al., 1981), were the first chemicals for which enhanced degradation in field soils was known to contribute to lack of expected performance. A number of different classes of pesticides are known to exhibit enhanced degradation in soils that were treated previously with these chemicals (Racke and Coats, 1990), including phenoxyalkanoic acids, carbamates, organophosphorus compounds, acetamides, anilides, and dicarboximides, among others. Microorganisms are responsible for enhanced degradation (Racke and Coats, 1990). The number of pesticides known to be affected by enhanced degradation continues to increase. As few as one field application of a pesticide may result in enhanced degradation. Ou (1991) found that degradation of the insecticide-nematicide fenamiphos was enhanced in a Florida sandy field soil treated with the chemical only once.

Degradation of 1,3-D in Soil

Both cis- and trans-1,3-D in soil are initially hydrolyzed to corresponding cis- and trans-3-chloroallyl alcohol (3-CAA) (Roberts and Stoydin, 1976), which in turn are oxidized to the corresponding cis- and trans-3-chloroacrylic acid (Fig. 1). The two acrylic acids are degraded to simple aliphatic carboxylic acids such as propionic acid, acetic acid, and succinic acid (Barnekow et al., 1995; DowElanco, 1996), which are then mineralized to final oxidation products, CO_2 and H_2O (Ou, 1989). Hydrolysis of 1,3-D to 3-CAA was considered by Roberts and Stoydin (1976) to be chemical, and subsequent steps of the degradation were thought to be biological. Degradation rates of cis- and trans-1,3-D in soils in the Netherlands were found to be similar kinetically (Leistra et al., 1991; Van der Pas and Leistra, 1987; Van Dijk, 1974, 1980). Half-life values of cis-1,3-D in these soils under laboratory conditions at 10, 15, and 20 °C ranged from 16 to 46, 7 to 33, and 3 to 19 days, respectively, whereas half-life values of trans-1,3-D under the same conditions ranged from 17 to 47, 4 to 32, and 3 to 15 days, respectively (Leistra et al., 1991; Van Dijk, 1974, 1980). Since cis- and trans-1,3-D are volatile, it is more difficult to accurately determine their degradation rates in soil than to determine the rates for nonvolatile pesticides. This may be why the reported half-life values in soils for each temperature were highly variable. These studies gave no information on the histories of 1,3-D applications to the field sites where soils were collected; therefore, it is not known whether previous applications of the chemical might have enhanced its degradation. Also, information on degradation rates of 1,3-D and its two isomers in soils under field conditions were not available. Since 1,3-D is volatile, it is very difficult, if not impossible, to accurately determine the degradation rates under field conditions.

Enhanced degradation of 1,3-D in soil was not known until 1989, when two studies (Lebbink et al., 1989; Smelt et al., 1989) reported the observation of rapid degradation of 1,3-D in soil suspensions and soils. Lebbink et al. (1989) found that 1,3-D in soil suspensions was rapidly degraded. Soil used in the suspensions was collected from a field site in the Netherlands that had been treated with 1,3-D annually for 12 years, and the suspensions were supplemented with inorganic and organic nutrients. Nematicidal efficacy in the field soil progressively declined with an increase in number of annual applications of 1,3-D over a period of 12 years with a 70% reduction of efficacy in killing potato cyst nematodes. This loss of nematicidal efficacy was attributed to the enhanced degradation of 1,3-D. Enhanced degradation of 1,3-D in some loamy soils in the Netherlands, with or without histories of previous field treatments of 1,3-D, was reported by Smelt et al. (1989). In both studies, individual degradation rates of cis- and trans-1,3-D were not determined. Further-

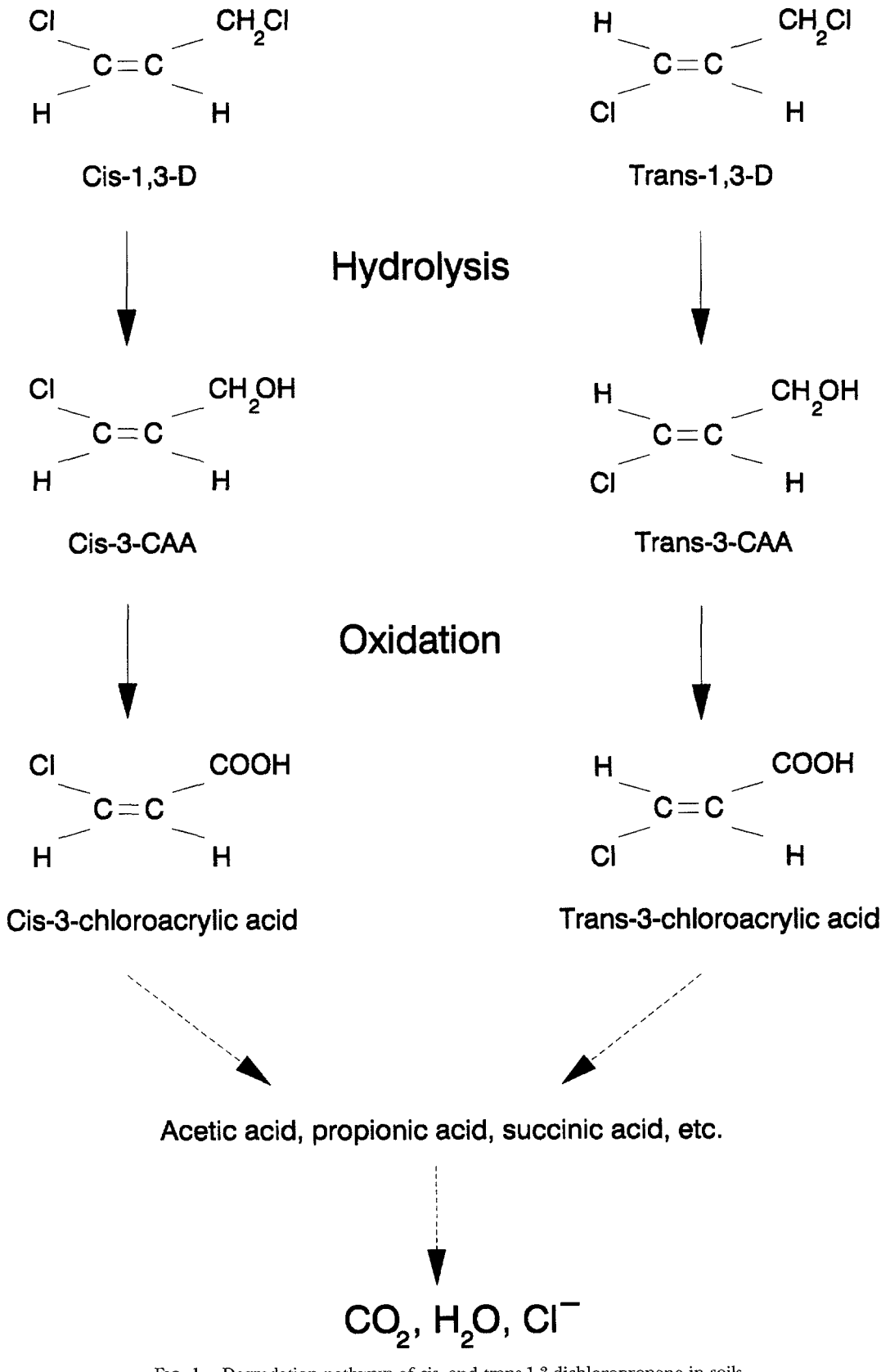

FIG. 1. Degradation pathways of cis- and trans-1,3-dichloropropene in soils.

more, corresponding untreated control sites were not established. Thus, it is questionable that enhanced degradation of 1,3-D truly occurred in the treated soils. Smelt et al. (1989) collected soil samples from 11 arable fields in three locations; 10 fields were from two locations reclaimed from a freshwater lake that itself reclaimed more than 100 years ago from the sea (G. Hoogeweg, pers. comm.). It appeared that so-called "enhanced degradation" reflected the intrinsic properties of the soils in which indigenous microorganisms in some fields had the capacity to degrade 1,3-D. Verhagen et al. (1995) demonstrated that extensive repeated applications of cis-1,3-D to microplots resulted in enhanced degradation of the isomer. After 1 year of treatment with cis-1,3-D repeated at 2-month intervals, degradation of this isomer in soils from the treated microplots was faster than in the corresponding soils from the untreated microplots. Neither 1,3-D nor trans-1,3-D was used in this study.

Enhanced degradation of 1,3-D in soil from a field site in Florida was observed by Ou et al. (1995). This site, which was planted with either peanut or tomato, had been treated with 1,3-D (Telone II) six times over the past 12 years at a rate of 55 to 115 liters/ha. A control site also was established near the treated site. Under laboratory conditions, not only was 1,3-D at an application rate of 40 μg/g degraded faster in treated soil than in untreated soil but trans-1,3-D in the treated soil was degraded faster than cis-1,3-D (Fig. 2). Application of 40 μg/g of 1,3-D to soil is equivalent to application of Telone II at 80 liters/ha. In contrast, the degradation rates of the two isomers in the untreated soil from the control site were statistically the same. Little or no trans-3-CAA

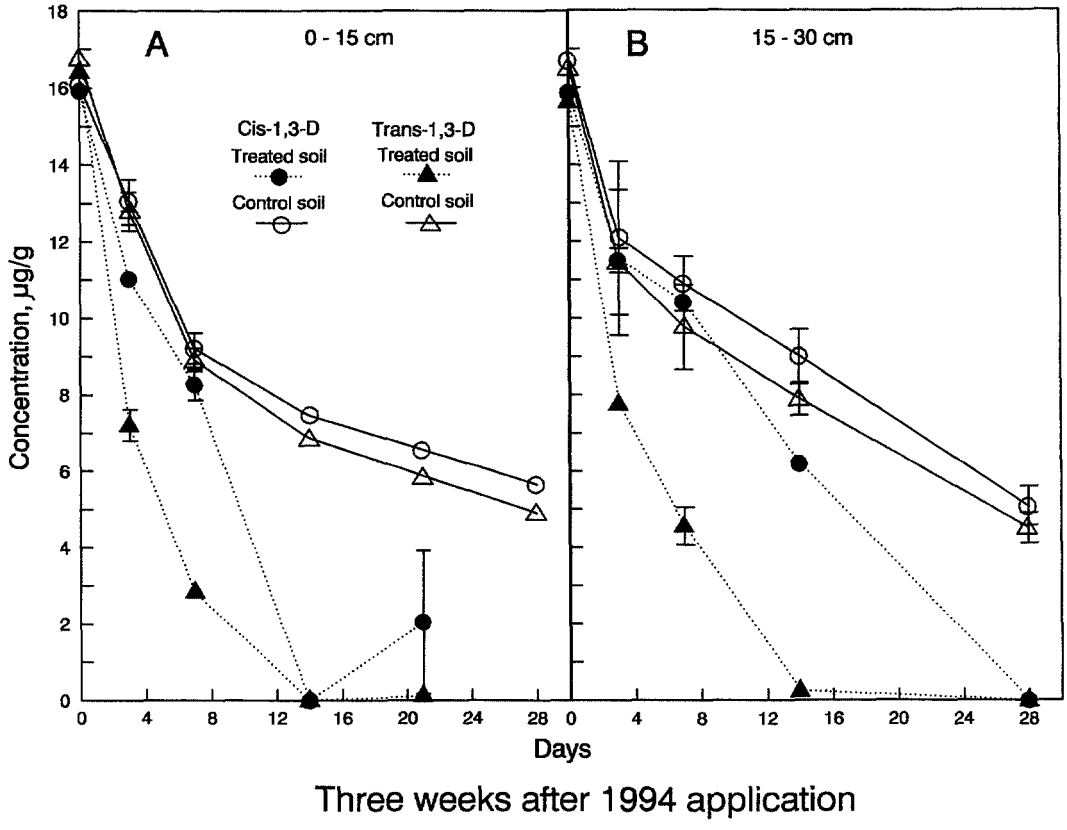

FIG. 2. Disappearance of cis- and trans-1,3-dichloropropene in surface (0- to 15-cm depth) and subsurface (15- to 30-cm depth) soil samples collected from the treated and untreated plots at a field site in Florida (Ou et al., 1995). Used with permission.

was detected in either treated or untreated soil, whereas substantial amounts of cis-3-CAA were formed, with more cis-3-CAA being detected in untreated soil than in treated soil.

Ou et al. (1995) concluded that biological hydrolysis was the main factor responsible for enhanced degradation of the trans-isomer in treated soil, and both biological and chemical hydrolysis contributed to the hydrolysis of the cis-isomer. Both cis- and trans-1,3-D in untreated soil were initially degraded mainly by chemical hydrolysis. 1,3-Dichloropropene in aqueous media was subject to rapid chemical hydrolysis (McCall, 1987), and the hydrolysis rate depended on temperature but was independent of pH at each temperature. At 20 °C and 30 °C, the half-life values for 1,3-D in water were 11.3 and 3.1 days, respectively, whereas the half-life values for cis- and trans-1,3-D in the treated surface soil (0 to 15 cm deep) at 25 °C were 8 and 3 days, respectively. In order for chemical hydrolysis to occur in soil, volatilized 1,3-D must be dissolved in soil solution. Hence, hydrolysis rates for 1,3-D in soils should be lower than in aqueous media. 1,3-Dichloropropene in soil is distributed into three phases: soil solution, sorbed to soil surfaces, and vapor phase. Since microorganisms degrade pesticides only in soil solution (Ogram et al., 1985), the sorbed 1,3-D must desorb into soil solution and the gas phase 1,3-D must be dissolved into the soil solution for degradation by microorganisms to occur. As a result, it is difficult to estimate the exact contribution of biological degradation on the degradation of cis- and trans-1,3-D in enhanced soil.

Lebbink et al. (1989) isolated a strain of *Pseudomonas* sp. enriched from a soil solution collected from a 1,3-D-treated field site. This bacterial isolate completely degraded 1,3-D in 6 days or less; and 1,3-D was degraded much faster than in the same medium without the isolate. This isolate differentially degraded the two isomers, with the trans-isomer being degraded faster than the cis-isomer. These findings were in agreement with those of Ou et al. (1995) that trans-1,3-D in enhanced soil was degraded faster than cis-1,3-D and that one out of three bacterial isolates from the soil solution was able to degrade 1,3-D. This indicated that a large number of bacteria capable of degrading 1,3-D existed in the treated field site. Verhagen et al. (1995) isolated 15 bacteria capable of degrading cis-1,3-D from microplot soils that showed enhanced degradation. Six of the degraders harbored a 50- to 60-kb plasmid, and there was some evidence that this plasmid might be involved in the degradation of cis-1,3-D. It was not reported whether these bacteria also could degrade trans-1,3-D. The first step of degradation by the six isolates appeared to be biological hydrolysis of cis-1,3-D to cis-3-CAA. Cis-3-chloroallyl alcohol was rapidly formed initially and then rapidly disappeared, indicating that these isolates also were capable of degrading cis-3-CAA. Ou (1989) enriched a mixed bacterial culture from a Florida sandy soil that was capable of mineralizing ^{14}C-1,3-D (an equal mixture of cis- and trans-^{14}C-1,3-D) to $^{14}CO_2$.

Degradation of Methyl Bromide in Soil

Since MeBr is a gas, degradation rates of MeBr in field soils are not known. It is a general practice that, immediately after the injection of MeBr, the fumigated soil is covered with polyethylene mulch, which may remain in place until the crop cycle is completed. Under such conditions, after 4 to 7 days of the injection little or no MeBr was found to emit into the atmosphere (Reible, 1994; Yagi et al., 1993, 1995; Yates et al., 1996). Within these periods, MeBr was either degraded in soil or volatilized into the atmosphere. Methyl bromide in soil is considered to be mainly degraded chemically, by chemical hydrolysis and methylation (Fig. 3) (Gan et al., 1994). Microorganisms, especially bacteria, also may be involved in the degradation of MeBr in soil (Short et al., 1995).

Microorganisms capable of utilizing short-chained halogenated hydrocarbons, including MeBr, have not been isolated from any environmental samples, including soils and

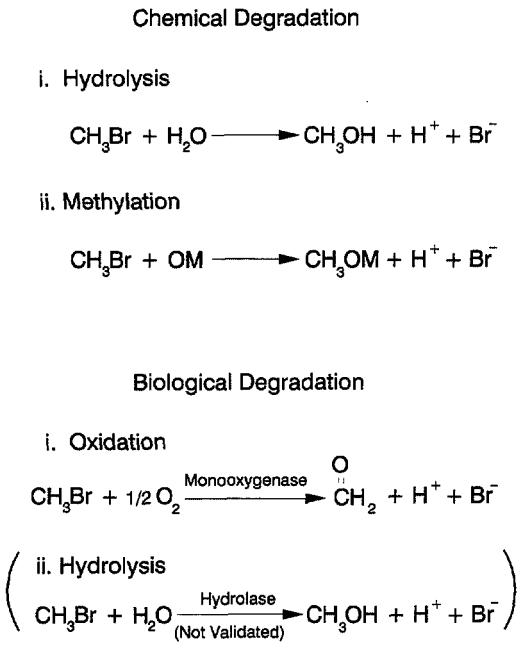

FIG. 3. Degradation pathways of methyl bromide in soils.

water. However, some bacteria that produce monooxygenases or dioxygenases such as methane monooxygenase, ammonia monooxygenase, or toluene dioxygenase for oxidation of the respective primary substrate (methane, ammonia, or toluene) also cooxidize some short-chained halogenated and nonhalogenated hydrocarbons, including trichloroethylene (TCE), chloroform, MeBr, ethene, propene, and others (Wackett, 1995). Rasche et al. (1990) were the first to demonstrate that two strains of soil ammonia oxidation bacteria, *Nitrosomonas europaea* and *Nitrosolobus multiformis,* had the capacity to cooxidize MeBr to formaldehyde and bromide ion (Fig 3). Oremland et al. (1994) subsequently showed that a strain of methane oxidation bacterium, *Methylococcus capsullatus,* had the capacity to mineralize ^{14}C-MeBr to $^{14}CO_2$.

Enhanced degradation of MeBr in soils from field sites that have been treated repeatedly with MeBr has not been reported. However, degradation of MeBr was greatly stimulated in methanotrophic soils (Oremland et al., 1994). At an application rate of 1,000 μg/g soil, MeBr in soils under aerobic conditions completely disappeared in 40 to 90 hours. Degradation of MeBr in methanotrophic soils is an oxidation process. At a low application rate (10 μg/g soil), MeBr in methanotrophic soils under aerobic conditions completely disappeared in 5 hours, whereas under anaerobic conditions, MeBr slowly disappeared, most likely from chemical degradation. In a recent study, Ou (1997) found that MeBr applied at 20 μg/g soil in a Florida sandy soil that had been treated with methane continuously for 1 month was completely degraded in 2 hours, and, at 50 μg/g soil, MeBr in the same soil was completely degraded in 1 to 3 days. Even though methanotrophic soils have a high capacity to degrade MeBr, the majority of agricultural soils are not methanotrophic and therefore are low in methane oxidation activity, with the exception of organic soils and former landfill sites. Thus, methane oxidation bacteria in agricultural soils likely contribute little toward the biological degradation of MeBr.

Ou et al. (1997) conjectured that when applied to soil for crop production, some ammonia compounds, such as ammonium sulfate found in nitrogen fertilizers, should stimulate soil-nitrifying activity. This, in turn, may stimulate the degradation of MeBr through the activity of ammonia monooxygenases produced during the oxidation of ammonia by nitrifying bacteria (Rasche et al., 1990). Methyl bromide in ammonium sulfate-treated Florida sandy soils, especially in a treated limed soil (pH 7.7), was initially degraded more rapidly than in untreated soil (Ou et al., 1997). Methyl bromide at an application rate of 20 μg/g soil in treated limed soil was degraded more rapidly over the first 5 to 7 days than in untreated limed soil (Fig. 4). Methyl bromide in autoclaved soil was degraded more slowly than in treated lime and untreated limed soils, indicating that biological degradation enhances the initial disappearance of MeBr in limed, treated or untreated soil. Since ammonia oxidation bacteria oxidize only the molecular form of ammonia, it would be expected that the increase in soil pH to 7.7 would increase the oxidation rate of ammo-

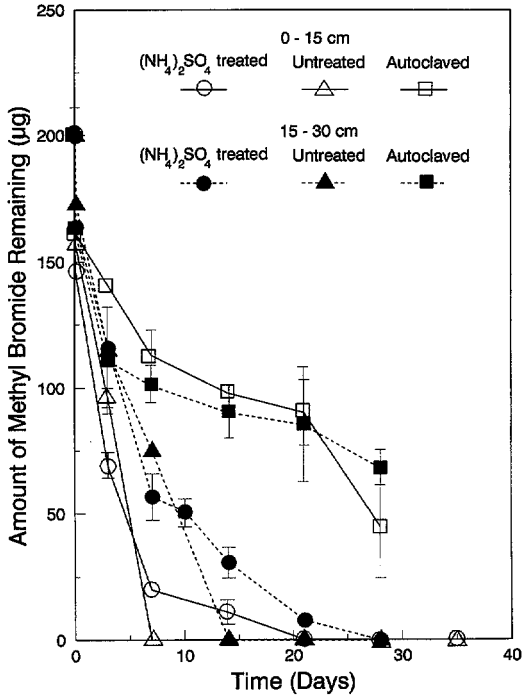

FIG. 4. Disappearance of methyl bromide in surface (0- to 15-cm depth) and subsurface (15- to 30-cm depth) Arredondo soil treated with MeBr (20 µg/g soil). Soil samples were limed and treated with $(NH_4)_2SO_4$, limed but not treated with $(NH_4)_2SO_4$, or limed and autoclaved (Ou et al., 1997). Used with permission.

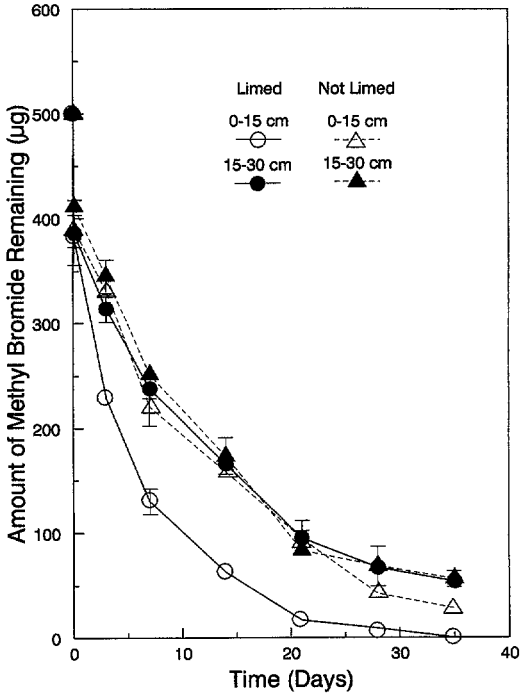

FIG. 5. Disappearance of methyl bromide in $(NH_4)_2SO_4$-treated surface (0- to 15-cm depth) and subsurface (15- to 30-cm depth) Arredondo soil. Soil samples (10 g) were treated with 500 µg of MeBr (50 µg/g soil) and were either limed or unlimed (Ou et al., 1997). Used with permission.

nia resulting in an increase of the oxidation rate of MeBr. Ou et al. (1997) also found that MeBr applied at a rate of 50 µg/g soil in limed, treated soil was initially degraded faster than in unlimed, treated soil (Fig. 5).

Based on information from field studies (Reible, 1994; Yagi et al., 1993, 1995; Yates et al., 1996), little or no MeBr volatilized into the atmosphere 5 to 7 days after field injection of MeBr. Therefore, in order to have an effective reduction of MeBr from volatilization into the atmosphere, biological stimulation of MeBr degradation in soil should occur within 5 to 7 days.

Both MeBr and 1,3-D are short-chained halogenated hydrocarbons, and MeBr has been demonstrated to be rapidly degraded in methanotrophic soils. However, degradation of 1,3-D appeared not to be stimulated in a Florida methanotrophic soil (Chung and Ou, unpubl.), even though such soils rapidly degraded MeBr (Ou et al., 1997). Biological hydrolysis also may be involved in the degradation of MeBr in soil (Fig. 3), but its involvement has yet to be proven. In addition to ammonia- and methane-monooxygenases, there are other mono- and dioxygenases involved in the initial step of degradation of some aromatic organic chemicals such as phenol, toluene, and 2,4-D. Phenol and toluene are toxic chemicals and are not suitable for applying to soil to stimulate MeBr. In contrast, the herbicide 2,4-D is commonly used in agriculture for control of broad-leaf weeds (Anonymous, 1967). One of the initial steps of 2,4-D degradation involves 2,4-dichlorophenol monooxygenase. This monooxygenase can cooxidize TCE (Harker and Kim, 1990). Application of 2,4-D may stimulate the degradation of MeBr in soil. Another scenario for stimulating MeBr degradation involves soil organic matter. Soil organic matter is rich in phenolic constituents (Stevenson, 1982),

and mono- or di-oxygenases responsible for degradation of the phenolic constituents also may degrade MeBr. Soils rich in organic matter, such as an organic soil, may degrade MeBr more rapidly than soils poor in organic matter.

Conclusions

Cis- and trans-1,3-D in soils are initially degraded through hydrolysis to cis- and trans-3-CAA, which are then oxidized to cis- and trans-3-chloroacrylic acid, then to aliphatic carboxylic acids such as propionic acid or acetic acid, and eventually to CO_2 and H_2O. Repeated field applications of 1,3-D resulted in enhanced degradation of 1,3-D in a Florida field soil, with trans-1,3-D being degraded more rapidly than cis-1,3-D. Degradation rates of cis- and trans-1,3-D in unenhanced soils were the same. In enhanced soil, biological hydrolysis is the main factor in the initial degradation of 1,3-D, especially trans-1,3-D, to 3-CAA, while chemical hydrolysis is responsible for initial degradation in unenhanced soils. Enhanced degradation may reduce the nematicidal efficacy of 1,3-D.

At present, it is not known whether repeated field applications of MeBr will induce enhanced degradation of the chemical in soil. However, it is known that MeBr degradation in soils is stimulated by some soil bacteria that produce certain monooxygenases, such as methane- and ammonia-monooxygenase, produced by methanotrophic bacteria and ammonia oxidation bacteria during the oxidation of their primary substrates, methane and ammonia. Under such conditions, MeBr is cooxidized to formaldehyde and bromide ion. Therefore, in addition to chemical degradation by means of chemical hydrolysis, biological oxidation also occurs in soils. Nitrifying bacteria are abundant in soils in the root rhizosphere, especially in agricultural soils, and are likely responsible for the biological degradation of MeBr. Although it has not been validated, biological hydrolysis of MeBr by soil bacteria also may be a factor in its degradation.

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