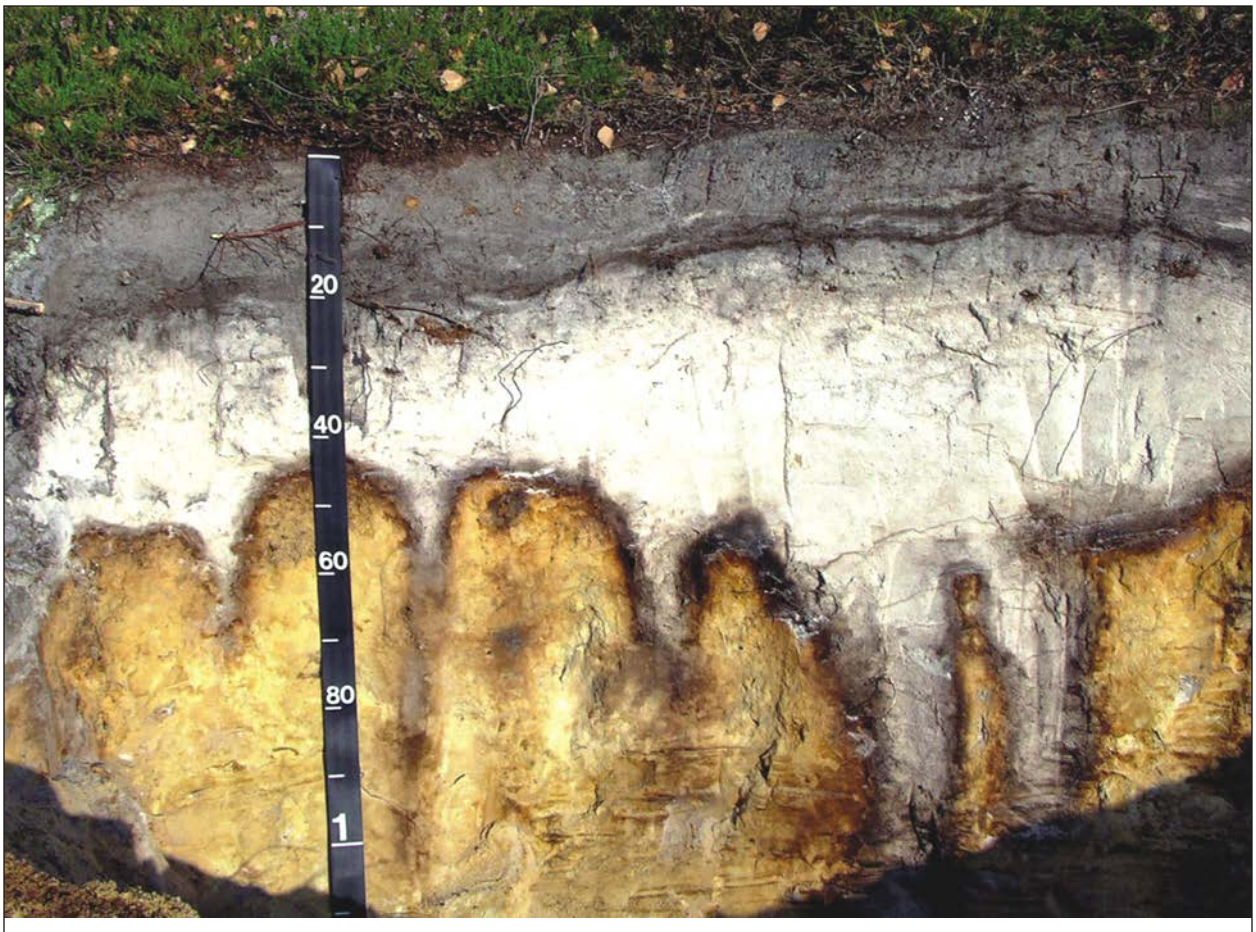

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Effects of Highly Volatile Organochlorine Solvents on Soil Respiration and Microbial Biomass

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Abstract: The effects of highly volatile organochlorine solvents (1,1,1-trichloroethane, TCET; trichloroethylene, TCE and tetrachloroethylene, PCE) on soil respiration and soil biomass were investigated using volcanic ash soil and gray lowland soil with different fertilizations. All the solvents significantly inhibited the activity and decreased the biomass under the sealed conditions 10 to 50 mg gds^{-1} solvents added. Under the upland and flooded conditions no significant difference between the solvents was observed. Taking into account the solubility of the solvents (TCET>TCE>>PCE) in water, the toxicity would be an order of PCE>TCE>TCET. Buffer ability, resulted from absorption of solvents on organic matters etc., was higher when the soil with organic manure fertilized was used. In addition, the effects were smaller on volcanic ash soil.

Key words: Soil respiration, soil biomass, 1,1,1-trichloroethane, trichloroethylene, tetrachloroethylene

Introduction

Volatile organic compounds, especially chlorinated aliphatic hydrocarbons such as 1,1,1-trichloroethane (TCET), 1,1,2-trichloroethylene (TCE) and tetrachloroethylene (PCE), are of major environmental concern (Cheremisinoff, 2001). Significant quantities of these compounds are detected in ground waters, especially around factories and laundries (Ohde and Bierod, 1989; Steve and Dale, 2005). As these compounds are suspected carcinogens or mutagens and potentially toxic to both human and microorganisms (Bouwer and McCarty, 1983), the movement of these polluted ground waters to agricultural fields and pollution of soil have been serious problems. Although degradation of these solvents and bioremediation of the soils polluted with them have been widely investigated (Lee *et al*., 1998; Fiedler and Lau, 1998; Okubo and Yagi, 1998; Middeldorp *et al*., 1998; Borch *et al*., 2003), information regarding their effects on soil microorganisms is very limited. To date, only the effects on soil enzyme activities (Kanazawa and Filip, 1986, 1987; Neumayr, 1981; Fuller *et al*., 1997) and the microbial biomass (Fuller *et al*., 1997; Skoda *et al*., 1984) have been reported. In the preliminary studies, application of 10 μg gds^{-1} of solvents inhibited all the enzymes tested (Kanazawa and Filip, 1986). As a continuation of these works, we began to study other biological

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effects of these solvents on soils, such as volcanic ash soil and gray lowland soil, typical representatives of Japanese arable soils. We first present herein the studies on the effects on soil respiration and microbial biomass.

Materials and Methods

This study was conducted at Faculty of Agriculture, the University of Tokyo from 1987 to 1990.

Soils

Soils of two types were collected. Properties of the soils are listed in Table 1. One was the plow layer of an upland field consisting of a volcanic ash soil of the Kanto loam type [FAO/UNESCO: Humic Andosols, US Soil Taxonomy: Typic (Lithic) Dystrandepts] located at the Tama farm of the University of Tokyo in Tanashi-City (Tokyo metropolitan area). Soil samples from three plots, one without fertilizer (abbreviated to NF), one plot with application of chemical fertilizer (nitrogen, 12 kg ha^{-1}; magnesium phosphate, 300 kg ha^{-1}, annually; abbreviated to CF) and one plot with farmyard manure+chemical fertilizer (farmyard manure, 6 t ha^{-1}; nitrogen, 12 kg ha^{-1}; fused magnesium phosphate, 300 kg ha^{-1}, annually; abbreviated to OM). Another was the plow layer of an experimental paddy field consisting of gray lowland soil (FAO/UNESCO: Eutric Fluvisols, US Soil Taxonomy: Typic Hydraquents) located at the Agricultural Research Center in Konosu-City (Saitama Prefecture). Soil samples were taken from three plots, one without fertilizer (abbreviated as NF), one plot with application of chemical fertilizer (N-P-K, 105-22-83 kg ha^{-1}, annually; abbreviated as CF) and one plot with farmyard manure+chemical fertilizer (organic manure, 12 t ha^{-1}; soybean cake, 600 kg ha^{-1}; P-K, 11-21 kg ha^{-1}, annually; abbreviated as OM). These soil samples were brought immediately to the laboratory, sieved (<2.0 mm) at field-moisture content and stored at 4°C. The pH was measured by a pH meter (soil:water ratio, 1:2.5). The total-C and -N contents in the soil were assayed with a C-N analyzer (Model MT 500, Yanagimoto MFG., Co., Ltd.). The moisture content was measured by drying at 105°C for 4 h.

Incubation

The incubation was carried out according to the literatures (Sayato and Ohsawa, 1983; Nakamuro, 1986) with a slight modification (vide infra).

Upland Conditions (Tanashi and Konosu)

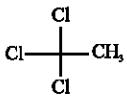
The pre-incubated soil sample (5 g dry weight) was placed in a 125 mL vial (total volume of gas phase was 0.12 L). To this was added each organochlorine solvent [0, 3, 10 or 50 mg g^{-1} dry soil (g ds)]. The experiment was run in duplicate for each sample. The vial was kept in the dark at 25°C.

Table 1: Properties of the soils used for the experiments

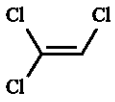
Site	Soil	Management	pH (H_2O)	pH (KCl)	Moisture (%)	Total C (%)	Total N (%)	C/N	Texture
		NF	6.3	5.1	29.0	2.1	0.19	11.2	LiC
Konosu	Paddy	CF	5.5	4.6	28.8	2.7	0.24	11.4	LiC
		OM	5.6	4.6	32.5	3.4	0.32	10.6	LiC
		NF	6.2	5.2	37.1	6.0	0.43	13.9	L
Tanashi	Upland	CF	6.0	5.3	37.9	6.0	0.45	13.3	L
		OM	6.1	5.6	38.6	7.3	0.55	13.2	L

NF, without fertilizer; CF, chemical fertilizer (nitrogen-fused magnesium phosphate, 12-300 kg/ha); OM, farmyard manure+chemical fertilizer (*farmyard manure, 6 t ha^{-1}; nitrogen-fused magnesium phosphate, 12-300 kg ha^{-1})

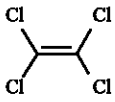
Table 2: Property of the organochlorine solvents



TCET
(1,1,1-trichloroethane)



TCE
(trichloroethylene)



PCE
(tetrachloroethylene)

Name	MW	Density	Solubility (mg L^{-1} H_2O)	Henry constant (L, atm/mol)
TCET	133.4	1.31	5500	4.9
TCE	131.4	1.46	1100	11.7
PCE	165.8	1.62	150	28.7

Table 3: Calculated concentration of the solvents in Konosu flooded conditions

Name	Distribution coefficient (gas/liq.)	Amount (mg g^{-1}ds)	Concentration (mg mL^{-1} H_2O)
TCET	3.7×10^{-2}	1	0.015
		5	0.073
		10	0.145
TCE	8.9×10^{-2}	1	0.005
		5	0.027
		10	0.055
PCE	1.7×10^{-2}	1	0.003
		5	0.013
		10	0.026

The gas phase of each sample was collected with a gas-tight syringe and CO_2 content was analyzed by a gas chromatography.

Paddy (Flooded) Conditions (Konosu)

The pre-incubated soil samples (7~11 g dry weight) and water (15 mL) were each placed in 125 mL vials. To this was added organochlorine solvent (0, 1, 10 mg g^{-1}ds). The vials were sealed as described for upland conditions and kept in the dark at 25°C. The gas phase of each sample was collected with a gas-tight syringe and the CO_2 concentration was analyzed by the gas chromatography. The experiment was run in duplicate for each sample. Properties and calculated concentration of organochlonine solvents in Konosu flooded conditions (Table 2 and 3)

Analysis

Analysis of Soil Respiration

Soil respiration was estimated by measuring the CO_2 concentration of gas phase of each vial. The gas phase (0.5 mL) was collected at 1, 2, 4 and 8 weeks after the incubation started. The same vials were used for each sampling. GC conditions: Shimadzu GC-3BT gas chromatograph; column, stainless spiral column (3 mm×2 m) with Porapak-Q (50-80 mesh); detector (TCD) temperature, 50°C; carrier gas, argon, 50 mL min^{-1}.

Analysis of Soil Microbial Biomass

Soil microbial biomass was measured as the ATP content according to the literatures (Jenkinson and Oades, 1979; Jenkinson *et al*., 1979)

Results

Figure 1 shows the result of soil respiration of Tanashi soil (upland conditions). Although the experiment was run only in duplicate, each value of the results was almost equal and reliable for

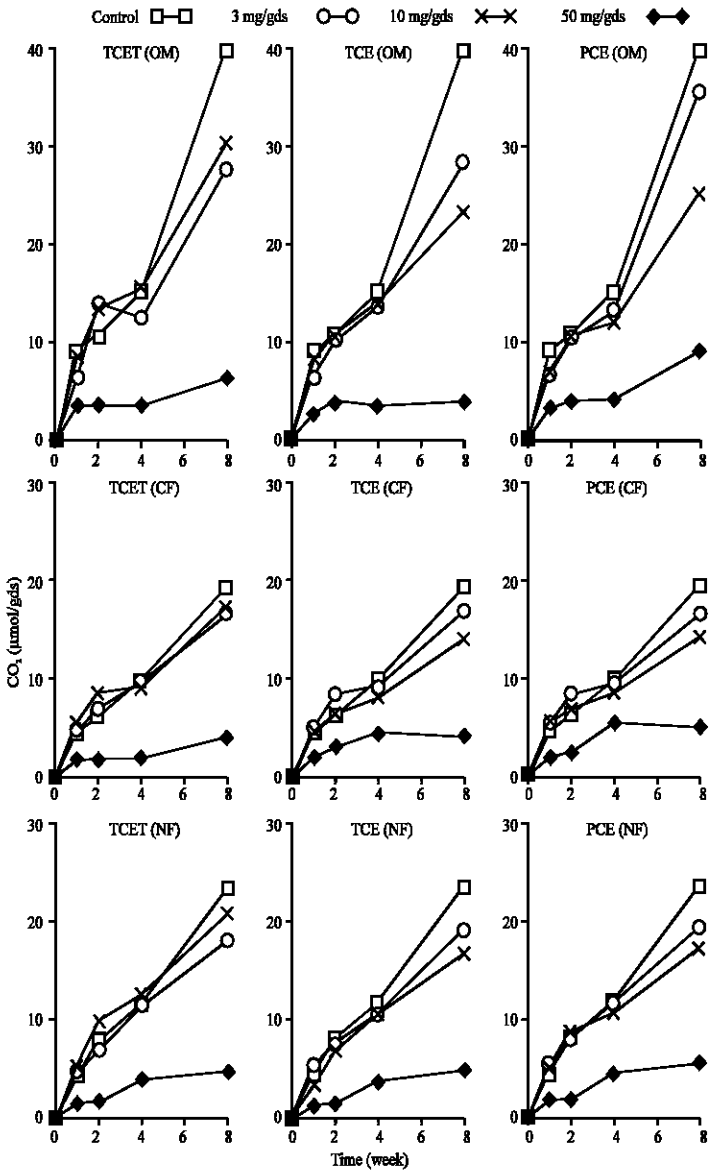


Fig. 1: Amounts of carbon dioxide in the headspace of each vial collected after 1, 2, 4 and 8 weeks of incubation (Tanashi, upland conditions). The experiments were run in duplicate for each sample NF: without fertilizer, CF: chemical fertilizer, OM: chemical fertilizer + farmyard manure

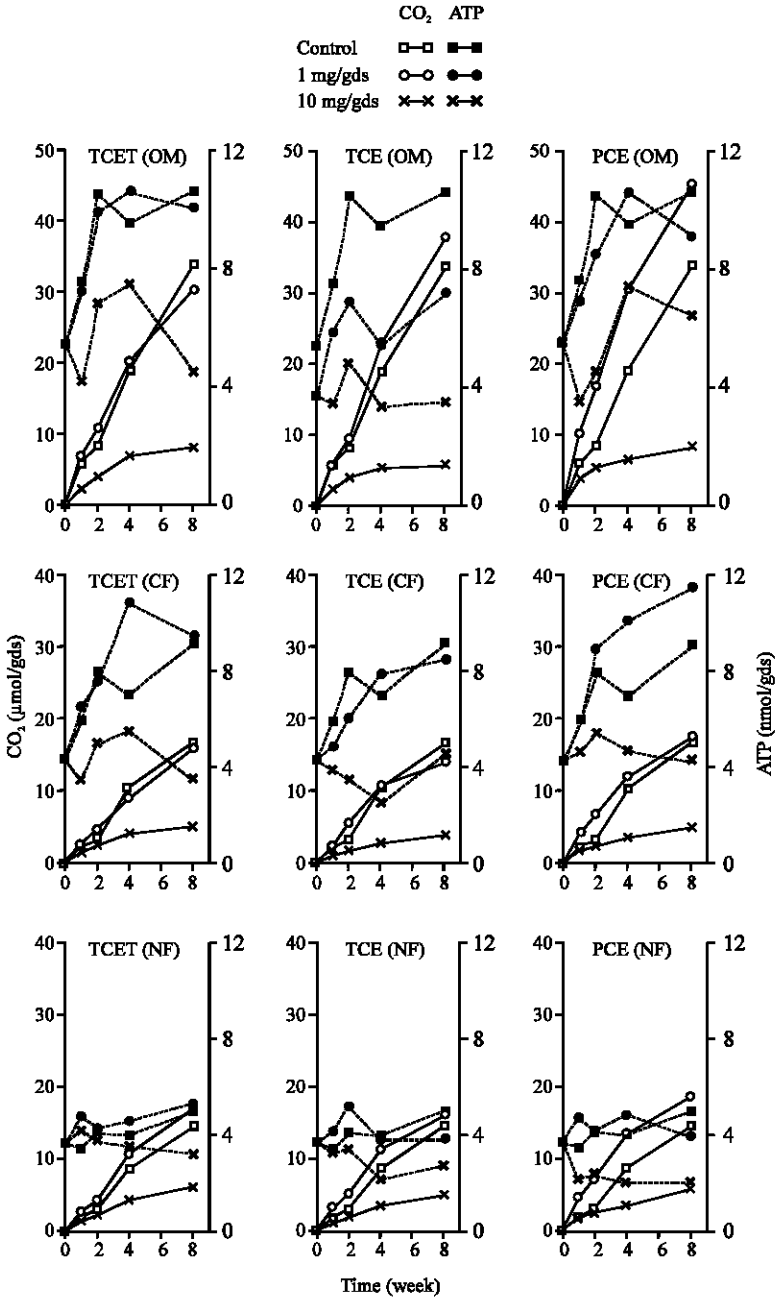


Fig. 2: Amounts of carbon dioxide in the headspace and ATP from the soil sample of each vial collected after 1, 2, 4 and 8 weeks of incubation (Konosu, upland conditions). The experiments were run in duplicate for each sample

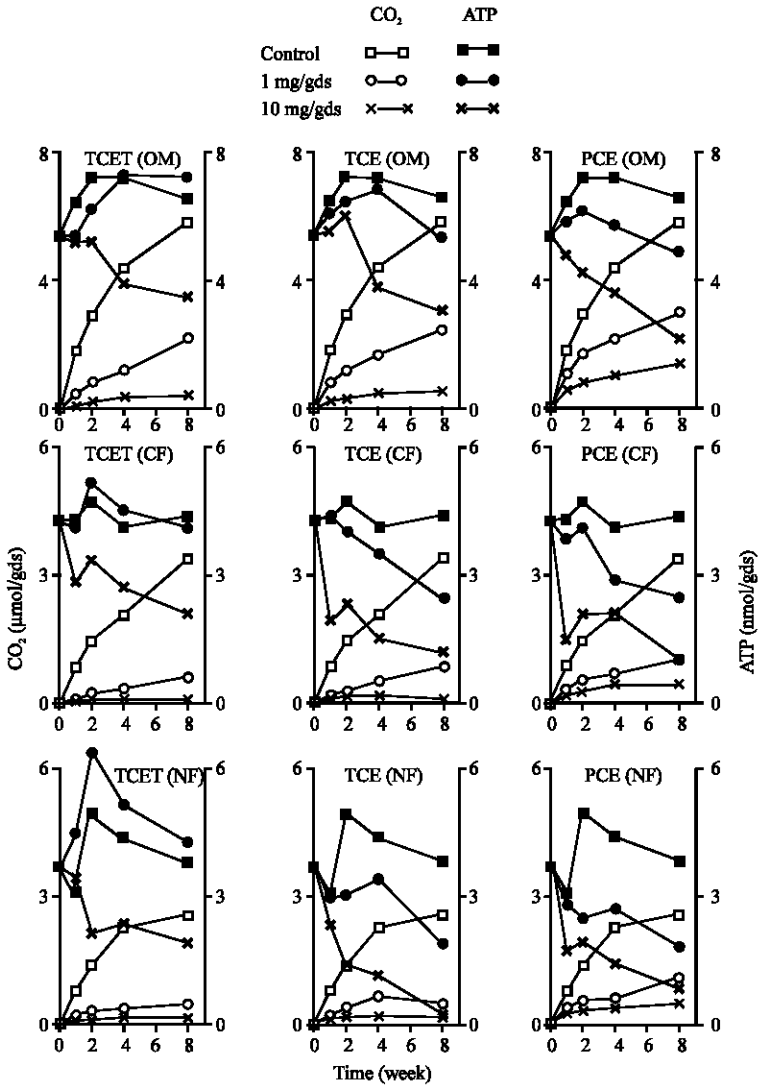


Fig. 3: Amounts of carbon dioxide in the headspace and ATP from the soil sample of each vial collected after 1, 2, 4 and 8 weeks of incubation (Konosu, flooded conditions). The experiments were run in duplicate for each sample

consideration. In each plot, addition of 50 mg g^{-1}ds of organochlorine solvent caused significant depression of CO_2 production and the soil respiration did not recover by 8 weeks. There was little difference between the effects of three solvents. Both the fertilization with organic manure (OM) or inorganic fertilizer (CF) did not alter the soil microbial activity.

Results on Konosu soil are shown in Fig. 2 (upland) and 3 (flooded). Under the upland conditions, the presence of 10 mg g^{-1}ds of the solvents resulted in a significant loss of CO_2 production.

All the solvents similarly affected almost all the soil. However, OM soil added with 1 mg of PCE was exceptional: the amount of CO_2 emitted from this soil exceeded that from the control soil. In addition, soil respirations was not suppressed by addition of all the solvents.

Under the flooded conditions, the inhibitory effect of solvents became stronger, even at the concentration of 1 mg g^{-1}ds: the evolved CO_2 decreased to 15 to 40% of controls. Similarly to the upland conditions, the buffer action of OM application was stronger and chemical fertilizer only was not effective as buffer. The inhibitory effects of the solvents were almost equal.

Soil microbial biomass indicated by the soil ATP concentrations are shown in Fig. 2 and 3. In both the upland and flooded conditions, decrease in ATP contents was considerable for 10 mg g^{-1}ds plots but not for 1 mg g^{-1}ds plots.

Discussion

Under the upland condition, 10 mg g^{-1}ds of all solvents did not effect so mush for the Tanashi soil but for the Konoshu soil, so the buffer action of Konosu soil seemed to be weaker than that of the Tanashi soil. This is probably due to the difference in clay minerals and organic compounds between the 2 soils. Volcanic ash soil is composed of allophane and contains highly humified humic substances, while gray lowland soil is composed of crystalline aluminosilicates and contains mildly humified humic substances with a low degree of humification. This suggests that the highly humified humic substances with hydrophobic nature has high ability of detoxication by adsorbing the organochlorine solvents. The high porosity of the allophanic volcanic acid may help the detoxication. Higher toxicity of the solvent under the flooded condition than under upland condition may be caused by inhibited adsorption of the solvents on the mildly humified humic substances.

The reason for that the ATP contents under flooded conditions did not decrease as the respiration did may be the time lag between the stop of soil respiration and the degradation of ATP after the death of microorganisms. Maintenance of ATP contents may help the recovery of soil activities.

The inhibitory effects of the three solvents on soil respiration and soil microbial biomass were almost equal even though their solubility in water is quite different. Thus, it is difficult to estimate the true toxicity of the solvents. The overall results showed that extremely large quantity of the solvents caused serious inhibition of soil activities. As these compounds are sometimes detected at over thousands times more than the permitted concentration around factories (Ohde and Bierod, 1989), the present data will be of help to anticipate the effects on heavily polluted agricultural fields with these solvents. Further studies of the effects on soil nitrogen metabolisms and number of soil microorganisms will be reported.

Acknowledgments

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