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Abiotic control on the dissipation of chlorpyrifos spiked in an Udic Ferrisol at a lower dosage

Ligang Wang^a, Wentao Jiao^b, Xin Jiang^a & Yongchao Liang^c

^a State Key Laboratory of Soil and Sustainable Agriculture,
Institute of Soil Science, Chinese Academy of Sciences, Nanjing
210008, China

^b Research Center for Eco-environmental Sciences, Chinese
Academy of Sciences, Beijing 100085, China

^c Institute of Agricultural Resources and Regional Planning, and
Key Laboratory of Plant Nutrition and Nutrient Cycling, Chinese
Academy of Agricultural Sciences, Beijing 100081, China

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Abiotic control on the dissipation of chlorpyrifos spiked in an Udic Ferrisol at a lower dosage

Ligang Wang^{a*}, Wentao Jiao^b, Xin Jiang^{a*} and Yongchao Liang^c

^aState Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing 210008, China; ^bResearch Center for Eco-environmental Sciences, Chinese Academy of Sciences, Beijing 100085, China; ^cInstitute of Agricultural Resources and Regional Planning, and Key Laboratory of Plant Nutrition and Nutrient Cycling, Chinese Academy of Agricultural Sciences, Beijing 100081, China

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The dissipation routes of chlorpyrifos spiked in an Udic Ferrisol at the dosage of 400 ng g⁻¹, which represents a residual level after an agricultural application, were studied in a laboratory, using the natural soil under three moisture regimes, sterilised, and organic matter-free soils in the dark at 20°C. The obtained results showed that higher soil moisture caused faster dissipation, and the abiotic and biotic contributions to the total dissipated amount of the applied chlorpyrifos in the soil accounted for 77.3–85.2% and 14.7–22.7%, respectively. In the sterilised soil, the contribution of organic matter to chlorpyrifos dissipation was noticed to account for 95% in the earlier two weeks, and was nearly equal to that of soil minerals two weeks after the incubation.

Keywords: abiotic factor; chlorpyrifos; dissipation; soil

1. Introduction

Chlorpyrifos (*O*, *O*-diethyl *O*-3, 5, 6-trichloro-2-pyridyl phosphorothionate) is an organophosphate insecticide that is widely used for pest control in agriculture and in turf and ornamental plants, to a lesser degree, for indoor use and soil applications to control termites.[1–4] Soil contamination by the pesticide can result from bulk handling in fields, containers rinsing and accidental release. A wide range of water and terrestrial ecosystems have been found to be contaminated with chlorpyrifos.[5–9] Chlorpyrifos' metabolism, cholinergic toxicity, and low reproductive toxicity have been reviewed by Richardson.[10] Chlorpyrifos may remain biologically active for periods ranging from 20 days to three months in soils.[11] Owing to relatively long persistence and ability to accumulate in living tissues, its potential risk to the health of humans and other organisms is of great concern.[12–14]

Half-lives of chlorpyrifos in soils determined under laboratory conditions vary greatly, ranging from less than 10 days to greater than 120 days.[15,16] In general, microorganisms demonstrate considerable capacity for the metabolism of many pesticides. Several attempts to isolate a chlorpyrifos-degrading microbial system by repeated treatments or enrichment of soils and other media with chlorpyrifos have failed until recent success by Singh et al.[17]

*Corresponding author. Emails: lgwang0827@aliyun.com; jiangxin@issas.ac.cn

The abiotic degradation processes of chlorpyrifos in soil include hydrolysis, oxidation, reduction, and photolysis on soil surface.[15] The major factors that influence abiotic dissipation of chlorpyrifos in soil involve moisture, pH, organic matter (OM), and clay content.[11,15,18,19] Although the degraded routes of pesticides in soil may be either abiotic or microbial in nature, the latter has received more attentions. There have been numerous investigations on microbial degradation of pesticide in soil, and several reviews are available.[15] In contrast, little information is available on the importance of abiotic dissipating processes of chlorpyrifos in soil. In this study, a series of experiments were conducted to test a hypothesis that the dissipation of chlorpyrifos spiked in soil at a lower dosage would be greatly affected by abiotic factors (soil moisture, soil minerals, and OM).

2. Materials and methods

2.1. Soil sample

The soil was sampled from 0–20 cm soil layer in Yintan eco-agricultural experimental station, Jiangxi Province in central south China. Peanuts were planted in field which had no applications of chlorpyrifos for at least two years. A soil sample with 30% moisture content at field moisture capacity (FMC, ratio of water retained in the soil after ample irrigation or heavy rain when the rate of downward movement has substantially decreased, usually one to three days after irrigation or rain, expressed as a percentage of water in soil.), pH 4.62 (1:1 soil–water ratio), OM 13.5 g kg⁻¹, CEC 18.2 cmol kg⁻¹, clay 45%, silt 34%, and sand 21%, was air-dried and sieved through a 2-mm mesh for laboratorial use. It was classified as an Udic Ferrisol which had developed from a typical quaternary red-earth parent material.

2.2. Chemicals

A HPLC grade chlorpyrifos with a purity of 99.5% was sourced from Dr. Ehrenstorfer GmbH (Augsburg, Germany). Solvents (*n*-hexane, acetone, and petroleum ether), and other reagents (sodium perborate, anhydrous sodium sulfate, and hydrochloric acid) were obtained from the Nanjing Chemical Reagent No. 1 Factory (Nanjing, China). Silica gel (0.063–0.200 mm particle size) was obtained from the Dalian Institute of Chemical Physics, Chinese Academy of Sciences. The preceding chemicals are all of analytical purity grade.

2.3. Soil sterilisation and OM elimination

The sterilised (ST) soil was prepared as 1 kg soil was sterilised twice with an autoclave for 50 min at 130°C. The OM-free soil was prepared as follows: 500 g soil was weighed into a beaker with a high base and was moistened with a small amount of distilled water (about 15 mL), then 6% H₂O₂ was added to oxidise soil OM, while the mixture was stirred with a glass rod and isoamylol was added to dispel gas bubbles. Finally, extra H₂O₂ was removed with further heating. The OM-free soil was sterilised with an autoclave to avoid biotic effect, prior to the incubation.

2.4. Dissipation experiment

Soil sample (5 g for each) was weighed into 50-mL colorimetric tubes, different soil–water contents (60%, 80%, and 100% FMC) were obtained by adding the calculated amounts of water. Soil moisture at 80% FMC (24% soil moisture content, measured on a gravimetric basis) was regarded

as an ideal level for crop growth and served as the control (CK). The soil (5 g, 4% moisture content) was evenly spiked with chlorpyrifos ($1 \mu\text{g/mL}$) in acetone (1.92 mL) at the level of 400 ng g^{-1} soil, which represented a normal residue in soil after chlorpyrifos application in agricultural production in China, according to the previous reports.[20–22] After the addition of chlorpyrifos, the tube was shaken to make blend uniform and allow acetone to evaporate. Then, the tube was plugged with the cotton wool and incubated in the dark at 20°C . During the incubation, distilled water, the amount of which was quantified by the weight difference between the initial sample and the incubated one, was added in an aseptic work bench once a week to maintain the constant soil moisture. Fifteen replicates under each moisture regime were conducted, three of which were used to test recoveries of chlorpyrifos in the soil, the remaining were used to analyse chlorpyrifos residue in turn on the 7th, 14th, 21st, and 28th day of the incubation with three replicates used at each interval, respectively. The dissipated amount of chlorpyrifos from the incubated soil was calculated by subtracting the residual concentration at end of the incubation from the initial level of 400 ng g^{-1} .

As the preceding incubation and test processes, the natural, ST, and OM-free soil (5 g for each) with chlorpyrifos spiked at 400 ng g^{-1} under 80% FMC regime were studied, respectively. The natural soil treatment was served as the control (CK).

2.5. Analysis of chlorpyrifos in the soil

Soil sample (5 g) in a 50-mL centrifuge tube was treated as follows: malathion ($100 \mu\text{L}$, $10 \mu\text{g mL}^{-1}$) was added as an internal standard, petroleum ether:acetone (25 mL, 2:1 v/v) as extractant was added to the tube and shaken for 2 h in an ultrasonic water-bath maintained at 20°C for extraction of chlorpyrifos. Then, the sample was centrifuged for 10 min at $1360.26 \times g$, separated into a 25-mL pear-shaped flask and pre-concentrated in a rotary evaporator to nearly 0.5 mL, which was then applied to the top of a solid phase extraction (SPE) cartridge. The preparation of SPE cartridge was detailed as Wang et al.[23] The flask was rinsed three times with 0.3 mL petroleum ether and the rinsate was also added to the top of the SPE column. The concentrated sample and rinsate were eluted with 15 mL petroleum ether: acetone (2:1). The eluate was collected in a 25-mL pear-shaped flask, then pre-concentrated in a rotary evaporator to nearly 1 mL, and transferred to a 1-mL measuring flask. The pre-concentrated sample was gently aired to dryness using nitrogen gas, and then dissolved with n-hexane and brought up to 1 mL volume, $1 \mu\text{L}$ of which was injected into the GC for analysis.

A Hewlett-Packard (HP, Santa Clarita, CA) 6890 GC with a nitrogen–phosphorus detector (NPD) for separation and quantitative analysis, and a HP-5MS 5% phenyl–methyl–siloxane capillary column (30 m length, 0.25 mm i.d., 0.25 μm film thickness) for gas chromatographic separation were used. N_2 at a rate of 1.5 mL min^{-1} was the carrier gas, hydrogen (4 mL min^{-1}) and air (60 mL min^{-1}) were combust gases for the NPD. The column head pressure was set at 85.7 kPa, the temperatures were programmed as follows: initial temperature 60°C for 1 min, then up to 140°C at $12^\circ\text{C min}^{-1}$, and finally up to 300°C at 8°C min^{-1} , whereupon it was held for 5 min. The injector and detector temperatures were set at 250 and 320°C , respectively. Retention time was used to identify the correct peak on the chromatogram.

2.6. Recovery of chlorpyrifos from the soil

Recovery tests were conducted by spiking chlorpyrifos at 400 ng g^{-1} in 5 g soil sample, with three replications for each treatment. The obtained average recoveries of chlorpyrifos from the natural soil under 60%, 80%, and 100% FMC regimes, OM-free and ST soil were $86.6 \pm 3.1\%$, $88.5 \pm$

Figure 1. Effect of soil moisture on dissipation of chlorpyrifos.

3.7%, $90.4 \pm 4.2\%$, $90.0 \pm 3.8\%$, and $88.6 \pm 4.3\%$, respectively. The recovery percentages of chlorpyrifos met the requirements of the study.

3. Results and discussion

3.1. Effect of soil moisture on the dissipation of chlorpyrifos in the soil

In comparison with the results of the control (80% FMC), the dissipated amount of chlorpyrifos under the lower moisture soil (60% FMC) was observed to decrease by 55.4%, 12.6%, and 6.13% on the 7th, 14th, and 21st day of the incubation, respectively. However, on the 28th day, both treatments caused the similar dissipated amount (Figure 1). Under the higher soil moisture (100% FMC) as compared to the control, the dissipated amount of chlorpyrifos in the soil was observed to decrease by 93.7% on the 7th day, but increase by 60.7%, 58.9%, and 72.9% on the 14th, 21st, and 28th day, respectively.

The results show that the dissipation of chlorpyrifos in the soil could be substantially changed through water addition. Most pesticides degraded faster in moist soil than in dry soil,[2,18,24] as those observed in our study. Previous research showed that microbial activity was enhanced in wetter soils. In addition, water molecules in a moist soil may also compete with some chemicals for binding sites, making the chemicals more available for biological degradation. Under 100% FMC, there was a lag period for dissipation of chlorpyrifos (about 10 days). After this lag, there was an accelerated dissipation phases, as shown in Figure 1. These might be attributed to the behaviours of anaerobic microorganisms (AM), because AM needed a period to gradually adapt to the anaerobic conditions, which was the lag time; and then AM would be propagating themselves, which brought growing population and consumption of nutrient materials that include the added chlorpyrifos. Although there was no direct evidence to support the fact of the anaerobic degradation of chlorpyrifos, a process of reductive dechlorination for chlorpyrifos had been proved by Abramowicz,[25] which implied that chlorpyrifos could be degraded by anaerobic microorganisms.

The dissipation kinetics of chlorpyrifos in the soil under three moisture levels fit well with the exponential decay pattern, with $r > 0.80$. The correlation results are summarised in Table 1. The calculated half-lives of chlorpyrifos in the soil are 32.7, 28.4, and 16.0 days under 60%, 80% and 100% FMC regimes, respectively, which correspond well with the previous reports.[15]

3.2. Dissipation of chlorpyrifos in ST soil

In comparison with the results of the control (the natural soil), the dissipated chlorpyrifos in the ST soil decreased by 17.3–29.4% (Figure 2), which was regarded as the abiotic dissipation amount (ADA). The abiotic dissipation process involved chemical degradation (hydrolysis, oxidation,

Table 1. Half-lives of chlorpyrifos in the soil under different moisture regimes.

Treatment	Dissipated rate(day ⁻¹)	R^2	Half-lives (day)
60%FMC	0.0212	0.8550	32.7
80% FMC(control)	0.0244	0.8463	28.4
100% FMC	0.0432	0.9690	16.0
ST (80% FMC)	0.0183	0.7900	37.9
OM-free ((80% FMC)	0.0062	0.7631	111.8

Notes: FMC denotes field moisture capacity; ST: sterilised soil; OM-free: organic matter-free soil.

Figure 2. Dissipation of chlorpyrifos in the sterilised soil.

Table 2. The biotic and abiotic contributions to the dissipation of chlorpyrifos in the soil.

	Incubation time/day			
	7	14	21	28
TDA, ng g ⁻¹	101	135	163	166
BDA, ng g ⁻¹	19.3	19.9	37	32.6
Percentage of BDA to TDA, %	19.1	14.7	22.7	19.6
ADA, ng g ⁻¹	82.1	115	126	134
Percentage of ADA to TDA, %	81.3	85.2	77.3	80.7
MDA, g g ⁻¹	3.61	6.19	63.3	71.3
Percentage of MDA to ADA, %	4.40	5.38	50.2	53.2
OMDA, ng g ⁻¹	78.5	109	62.7	62.7
Percentage of OMDA to ADA, %	95.6	94.8	49.8	46.8

Notes: TDA denotes the total dissipation amount, BDA denotes the biotic degraded amount, ADA denotes the abiotic dissipation amount, MDA denotes the mineral-causing dissipation amount, OMDA denotes the organic matter-causing dissipation amount.

reduction), bound residue, and gaseous escape. However, the dissipation arising from gaseous escape would be negligible in this study because the incubation was done in a closed tube. The dissipated amount of chlorpyrifos in the natural soil was regarded as the total dissipation amount (TDA) in respective incubation period. The biotic dissipation amount (BDA) of chlorpyrifos in the soil was obtained from the calculation with TDA subtracted ADA in the same incubation period. The biotic dissipation means an actual process of biotic degradation. As shown in Table 2, the ADA and BDA accounted for 77.3–85.2% and 14.7–22.7% of the TDA, respectively. These results indicated that the abiotic dissipation played a more important role than the biotic degradation route in this study.

Previous investigations had reported that microbial activities were more important in the dissipation of chlorpyrifos in soil.[11,18] However, several investigators had noted that little difference was observed in the dissipation rates of chlorpyrifos between sterile and microbially active soils and had concluded that microorganisms might not always play a significant role.[16,26] This discrepancy regarding biotic and abiotic contribution to chlorpyrifos dissipation in soil might be attributable to the differences in originally applied dosage of chemicals, soil type, microbial activity and so on. A lower dosage of organic pollutant fortified in soil might be not enough to stimulate microorganisms' ability to degrade pollutant, because they possibly ignored this intervention from a lower dosage of chemical, owing to sufficient carbon and nutrition sources from soil. Soil sterilisation process might alter some soil properties and affect soil adsorptive potential, and this alteration may change the dissipating ability of the soil to chlorpyrifos. In the future study, it would be essential to investigate how the sterilisation alter soil properties, and which sterilisation method would be optimum to reduce this alteration to original soil.

3.3. Dissipation of chlorpyrifos in OM-free soil

In comparison with the results of the control and the ST soil, the dissipated amounts of chlorpyrifos in the OM-free soil decreased significantly (Figure 2), a dissipation lag was observed in the OM-free treatment, which might result from a slow sorption mechanism and continued hydrolysis of soil minerals to chlorpyrifos. The ST soil was consisted of minerals and OM. Soil minerals solely remained after soil OM free. The dissipation amount in soil minerals (namely MDA) was shown in Table 2. Similarly, the dissipated amount due to OM (namely OMDA) can be obtained from the calculation with ADA subtracted MDA in respective incubation period. The function of soil OM to dissipation might involve adsorption and bonding reaction. The MDA and OMDA were

calculated to account for 4.4–5.38% and 94.8–95.6% of the ADA during the initial 14 days of the incubation, respectively. However, both had a similar contribution to the total abiotic dissipation 14 days after the incubation (Table 2).

4. Conclusions

The half-lives of chlorpyrifos in the natural soil ranged from 16 to 33 days. The dissipation rate of chlorpyrifos decreased under low soil moisture (60% FMC), but increased by 59–73% under 100% FMC over 2 weeks. This finding has the practical implications that chlorpyrifos residue in soil could be substantially changed through water irrigation. The abiotic contribution to the total dissipation of chlorpyrifos fortified in the soil accounted for 77–85% in this study, this illustrates the relative importance of the abiotic dissipation and might imply an advance in our understanding dissipation route of pesticide residue in soil after an agricultural application. In the ST soil, OM played a dominant role in dissipation of chlorpyrifos in the soil in the earlier 2 weeks, whereas soil mineral and OM had similar contributions to the abiotic dissipation of chlorpyrifos 2 weeks after the incubation.

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