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Isomer Selectivity in Aquatic Toxicity and Biodegradation of Cypermethrin

WEIPING LIU,† JAY J. GAN,*,† SANGJIN LEE,† AND INGEBORG WERNER‡

Department of Environmental Sciences, University of California, Riverside, California 92521, and Department of Anatomy, Physiology and Cell Biology, University of California, Davis, California 95616.

Synthetic pyrethroids (SPs) are widely used in both agricultural and urban regions for insect control. Unlike many other pesticides, SPs are chiral compounds consisting of stereoisomers. However, occurrence of isomer selectivity in environmental processes is poorly understood for SPs. We evaluated isomer selectivity in toxicity of cypermethrin (CP) to *Ceriodaphnia dubia* and in its biodegradation by microbial isolates and in sediment. Among the eight enantiomers, two enantiomers (*1R-cis-*α*S* and *1R-trans-*α*S*) were found to be toxic to *C. dubia*. Bacteria strains isolated from sediment selectively degraded CP diastereomers and enantiomers. The trans diastereomers were preferentially degraded over the cis diastereomers. Of the two active enantiomers, *1R-cis-*α*S* was degraded slower, whereas *1R-trans-*α*S* was degraded faster than the other stereoisomers. Similar isomer selectivity was observed during CP degradation in whole sediment. Since ecotoxicity is likely caused only by the biologically active enantiomers, knowledge on isomer selectivity may improve our understanding of the ecological risks of CP and analogous SPs.

KEYWORDS: Isomer selectivity; enantioselectivity; cypermethrin; synthetic pyrethroids; biodegradation.

INTRODUCTION

Synthetic pyrethroid (SP) insecticides have been used for over two decades to control insects in crop production and around households. The historical concern about the use of SP compounds is their potential toxicity to fish and aquatic invertebrates (*1*−*4*). This concern had attracted renewed attention as recent studies showed potential transport of SP residues via runoff to surface streams (*5*−*7*). Once in surface water systems, SPs are expected to rapidly deposit to the sediment layer due to their strong adsorption potential (*2*, *8*, *9*). Therefore, the effect of SPs on surface water quality depends closely on their fate in the sediment phase, especially their persistence in the sediment that is controlled mainly by biodegradation.

All known SPs are chiral compounds that contain multiple stereoisomers (*10*). It is increasingly recognized that, for a chiral compound, biologically mediated environmental processes, including biodegradation and bioaccumulation, are isomer selective (*11*−*15*). Insecticidal activity of SPs is known to vary greatly among the different stereoisomers, and isomer-enriched products are intentionally manufactured to achieve greater pest control efficacy (*10*, *16*). However, isomer selectivity in environmental processes, e.g., aquatic toxicity and biodegradation, has been largely ignored so far in the risk assessment for SPs (*10*, *17*). The lack of study may be attributed to analytical difficulties in separating and identifying SP stereoisomers.

We recently developed analytical methods that allow separation of the diastereomers and *cis*-enantiomers in SPs (*18*). In this study, we characterized isomer selectivity of cypermethrin (CP) [(*RS*)-α-cyano-3-phenoxybenzyl (*1RS*)-*cis*−*trans*- 3-(2,2-dichlorovinyl)-1,1-dimethyl-cyclopropanecarboxylate] in toxicity to a commonly used freshwater indicator invertebrate *Ceriodaphnia dubia* and in biodegradation. Cypermethrin contains two chiral carbons in the cyclopropyl ring and one chiral position at the α-cyano carbon (**Figure 1**). Therefore, nonenriched formulations of CP consists of eight stereoisomers that form two cis diastereomers, *1S-cis-*α*S* + *1R-cis-*α*R* and *1R-cis-*α*S* + *1S-cis-*α*R*, and two trans diastereomers, *1R-trans-*α*R* + *1S-trans-*α*S* and *1S-trans-*α*R* + *1R-trans-*α*S* (**Figure 1**). In CP, it is known that *1R-cis-*α*S** and *1R-trans-*α*S** are the only isomers with insecticidal activity ("*" is used hereafter to indicate insecticidal activity) (*4*). These enantiomers are intentionally enriched in various commercial formulations of CP, including α-CP, *β*-CP, and *θ*-CP, to achieve improved insecticidal efficacy (*10*). Isomer selectivity of CP was previously studied only during degradation in soil (*17*), and no information exists on isomer selectivity in relation to aquatic toxicity or biodegradation in sediments. Findings from this study may be used to better understand the ecotoxicological effects of CP in aquatic systems.

MATERIALS AND METHODS

Chemicals and Isomer Makeup. Nonenriched, racemically mixed CP (CP, 98%) and *β*-CP (86%, enriched in *1R-cis-*α*S** + *1S-cis-*α*R* and *1S-trans-*α*R* + *1R-trans-*α*S**) were purchased from Chem Service (West Chester, PA). The isomer-enriched formulations α-CP (96%,

* To whom correspondence should be addressed. E-mail: jgan@ucr.edu.
† University of California, Riverside.
‡ University of California, Davis.

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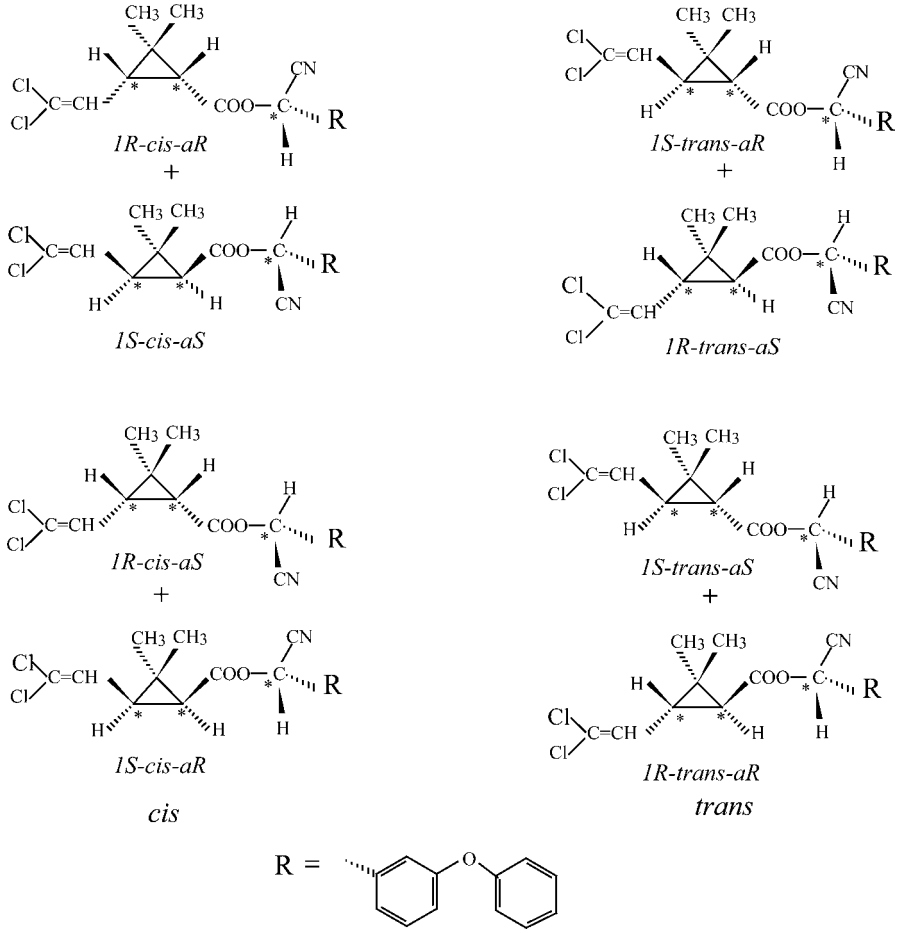

Figure 1. Structure of cypermethrin showing asymmetric carbon positions at 1C, 3C, and αC.

enriched in *1R-cis*-α*S** + *1S-cis*-α*R*) and *θ*-CP (99%, enriched in *1R-cis*-α*S**) were provided by FMC (Princeton, PA). Before use, samples of CP, α-CP, *β*-CP, and *θ*-CP were analyzed by gas chromatography (GC) to determine the relative content of each diastereomer and enantiomer. The stereoisomer content was calculated based on relative peak areas, assuming that each stereoisomer gave the same GC response. Pesticide stock solutions were prepared in acetone at 1.0×10^6 μg L^{-1} due to the very low aqueous solubility of CP (0.2 mg L^{-1}).

Aquatic Toxicity Assay. Isomer selectivity of CP enantiomers was evaluated through a 96-h acute toxicity assay using *C. dubia*. The overall procedure for the test was similar to the guidelines given in the EPA methods for measuring acute toxicity of effluents and receiving waters (*19*). Briefly, spring water (Arrowhead) purchased from a local grocery store was used to prepare the reconstituted, moderately hard water (RMHW) by adding the suggested salts (*19*). Test solutions containing CP, α-CP, *β*-CP, and *θ*-CP at various concentrations were prepared by serial dilution from pesticide stock solutions using the RMHW. The highest acetone content in each vial was <0.03 μL. 15 mL of the prepared solutions were transferred to 20-mL borosilicate glass scintillation vials, and four replicates were prepared for each concentration level. A treatment with no pesticide but with the same amount of acetone was also prepared and used as the control. Five active *C. dubia* neonates aging <20 h were added into each vial. The test organisms were fed with yeast cerophylla trout chow and *Selenosturm* sp. (Aquatic Research Organisms, Hampton, NH) for 4 h prior to the exposure. All vials were monitored at 24-h intervals until reaching 96-h exposure. The concentration that caused 50% mortality of the test population, or LC50 (μg L^{-1}), was determined by probit analysis using ToxCalc (v5.0) (Tidepool Scientific Software, McKinleyville, CA).

Biodegradation Experiment. Isomer selectivity was further evaluated during degradation of CP by bacterial isolates and in whole sediment under aerobic conditions. Details for isolation and characterization of pyrethroid-degrading bacteria were given in a previous study (*20*). Briefly, a sample of sediment that was previously contaminated with bifenthrin and permethrin was collected from a nursery runoff channel and used as the bacteria source. Bacterial degraders were enriched by adding cyfluthrin [(*RS*)-α-cyano-4-fluro-3-phenoxybenzyl (*1RS*)-*cis*−*trans*-3-(2,2- dichlorovinyl)-1,1-dimethylcyclopropane-carboxylate] as the sole carbon source to the sediment in a mineral-salt-based solution medium. Cyfluthrin is structurally analogous to CP, and both compounds contain chiral carbons at the same positions. Three fast-growing bacteria strains were selected as inoculants for the biodegradation experiment. Identification of the selected bacteria was made using fatty acid profiling and DNA sequence analysis (*20*). The three bacterial degraders of CP were identified as *Vibrio hollisae*, *Burkholderia picketti*, and *Erwinia carotovora*, and were labeled herein as CF-3, CF-17, and CF-28, respectively. Mineral salt solution used in the biodegradation experiment contained the following ingredients on per liter basis: 0.5 g of K_2HPO_4, 0.5 g of $NaNO_3$, 0.2 g of $MgSO_4 \cdot 7H_2O$, trace $FeSO_4 \cdot 7H_2O$, and 15 g of agar. The solution was adjusted to pH 7.0 with NaOH solution and was then spiked with the pesticide stock solution to give a nominal CP concentration of 100 μg L^{-1}. The initial acetone concentration in the test solution was estimated to be 100 μg mL^{-1}. 20 mL of the pesticide solution were transferred to 50-mL amber glass vials and amended with 1.0 mL of enriched bacteria solution. The initial bacterial population densities in the inoculated vials were 8.3×10^7, 2.4×10^8, and 2.5×10^8 colony-forming unit (CFU) mL^{-1}, for CF-3, CF-17, and CF-28, respectively. A total of 24 sample vials were prepared for each bacteria-pesticide combination, and 8 noninoculated vials were included as the control treatment. All sample vials were continuously mixed on a mechanical shaker at low speed at room temperature. At different time intervals, triplicate sample vials from each treatment were removed and immediately extracted for pesticide residues. Briefly, 10 mL solution sample was transferred to a 250-mL glass separatory funnel and was manually mixed with 50 mL of ethyl acetate for 1 min. The same extraction procedure was repeated for two consecutive times, and the solvent extracts were combined. The ethyl acetate extract was

filtered through 50 g of anhydrous sodium sulfate to remove the residual water and then concentrated to near dryness on a vacuumed rotary evaporator at 60 °C. The pesticide residues were recovered in 4 mL of hexane-acetone (1:1, v/v), and an aliquot was transferred to a 2-mL autosampler vial for analysis by GC. The above extraction procedure gave >95% recovery for CP or CP isomers.

Degradation in Whole Sediment. Isomer selectivity in CP degradation was further evaluated by investigating changes in the isomer composition during degradation in whole sediment. The sediment sample was collected from a sedimentation pond at a nursery site in Irvine, CA, and contained 0.65% organic carbon and 5% clay. Chemical analysis showed that the sediment was free of CP residues. The sample was drained of free water and used without air-drying to preserve the original microbial activity. 50 g (dry weight equivalent) of the sediment were placed in a 250-mL glass flask and then immersed with 80 mL of deionized water. 50 μL of CP stock solution (1 mg mL^{-1}) was added to the sample flask and manually mixed. The initial CP concentration was thus 1.0 mg kg^{-1}. The sample flasks were loosely covered with aluminum foil and kept at room temperature (21 ± 2 °C). 5 g of sediment was removed at different times after the treatment and analyzed for concentrations of CP diastereomers and enantiomers. The sediment sample was mixed at high speed for 60 min with 20 mL of acetone-hexane (1:1, v/v) in a 50-mL Teflon centrifuge tube and then centrifuged at 1300 × g for 20 min. After the supernatant was decanted, the remaining sediment phase was extracted two additional times using fresh solvents. The extracts were combined, dried with 50 g of anhydrous sodium sulfate, and then concentrated to 5.0 mL on a vacuumed rotary evaporator at 45 °C. An aliquot was used for analysis of CP diastereomers and enantiomers by GC. The recovery of the above procedure was >90%.

Stereoisomer Separation and Analysis. The GC methods for separating and identifying CP diastereomers and enantiomers were described in detail elsewhere (*18*). Briefly, separation and analysis were carried out on an achiral column for CP diastereomers and on a chiral column for CP enantiomers. An Agilent 6890N GC with electron capture detector (ECD) was used for both achiral and enantioselective analyses. The temperature of ECD was 310 °C, and the detector makeup gas was N_2 (60 mL min^{-1}). The inlet temperature was 260 °C, and 1 μL was introduced in the splitless mode. The flow rate of the carrier gas (helium) was 1.5 mL min^{-1}. Achiral GC analysis was carried out on a 30 m × 0.25 mm × 0.25 μm HP-5MS column (cross-linked 5% diphenyl and 95% dimethyl-polysiloxane, Agilent, Wilmington, DE). The initial column temperature was 180 °C for 2 min, and ramped at 5 °C min^{-1} to 280 °C, followed by an isothermal hold at 280 °C until complete elution. Enantioselective GC analysis was carried out on a 30 m × 0.25 mm × 0.25 μm BGB-172 column (20% *tert*-butyldimethylsilyl-β-cyclodextrin dissolved in 15% diphenyl- and 85% dimethyl-polysiloxane, BGB Analytik, Adliswil, Switzerland). The column was initially held at 160 °C for 2 min, ramped at 1°C min^{-1} to 220 °C (first ramp), held at 220 °C for 60 min, ramped at 5 °C min^{-1} to 230 °C (second ramp), and then held at 230 °C until complete elution.

RESULTS AND DISCUSSION

Isomer Selectivity in *C. dubia* Toxicity. The isomer composition of CP and its isomer-enriched formulations α-CP, β-CP, and θ-CP was determined by comparing relative peak areas of the resolved peaks. All diastereomers were separated at the baseline on the achiral HP-5MS column (**Figure 2**a). On the enantioselective BGB-172 column, separation was achieved for all enantiomers from the cis diastereomers but not for enantiomers from the two trans diastereomers (**Figure 3**a). Assuming equal fractions of *1S-trans-*α*R* and *1R-trans-*α*S** in the unresolved trans diastereomer *1S-trans-*α*R* + *1R-trans-*α*S**, the content of insecticidally active enantiomers (sum of *1R-cis-*α*S** and *1R-trans-*α*S**) increased in the order CP (22.0%) < α-CP (41.4%) ≈ β-CP (41.2%) < θ-CP (83.0%) (**Table 1**).

The relative *C. dubia* toxicity of the different CP stereoisomers was inferred from the LC50 values measured for CP and the various isomer-enriched CP products (**Table 1**). The LC50

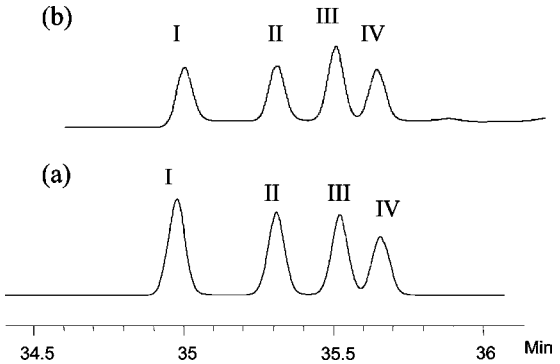

Figure 2. Achiral gas chromatograms of cypermethrin (CP). (a) Nonenriched CP formulation before degradation. (b) Nonenriched CP formulation after 5 h of biodegradation by CF-3 (*I* = *1R-cis-*α*R* +*1S-cis-*α*S*; II = *1R-trans-*α*R* + *1S-trans-*α*S*; III = *1R-cis-*α*S* + *1S-cis-*α*R*; IV = *1R-trans-*α*S* + *1S-trans-*α*R*).

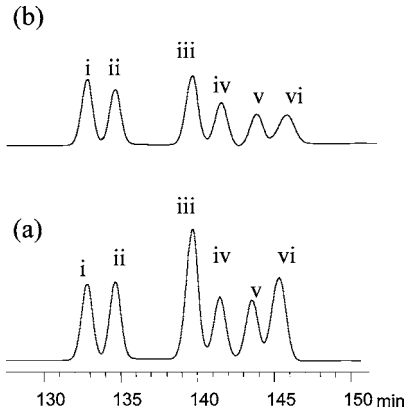

Figure 3. Chiral gas chromatograms of cypermethrin (CP). (a) Nonenriched CP formulation before degradation. (b) Nonenriched CP formulation after 5 h of biodegradation by CF-28 (*i* =*1R-cis-*α*R*; *ii* =*1S-cis-*α*S*; *iii* = *1R-trans-*α*R* + *1S-trans-*α*S*; *iv* = *1R-cis-*α*S*; *v* = *1S-cis-*α*R*; *vi* = *1R-trans-*α*S* + *1S-trans-*α*R*).

decreased concurrently as the content of insecticidally active enantiomers increased, in the order CP (0.889 μg L^{-1}) > α-CP (0.696 μg L^{-1}) ≈ β-CP (0.690 μg L^{-1}) > θ-CP (0.356 μg L^{-1}). Linear regression of the LC50 values against the content of insecticidally active enantiomers from **Table 1** showed close correlation (r^2 = 0.995), suggesting that the insecticidally active enantiomers were also responsible for the observed aquatic toxicity. Therefore, it may be concluded that only two of the four diastereomers, i.e., *1R-cis-*α*S** + *1S-cis-*α*R* and *1S-trans-*α*R* + *1R-trans-*α*S**, or two of the eight enantiomers, i.e., *1R-cis-*α*S** and *1R-trans-*α*S**, contributed to the observed *C. dubia* toxicity. This analysis suggests that enantioselectivity in *C. dubia* toxicity coincided with that in insecticidal activity for CP enantiomers (*4*). Leicht et al. (*16*) evaluated toxicity of cyfluthrin to *Daphnia magna* and various strains of *Lepidoptera* and also observed that the toxicity was consistently attributable to the *1R-cis-*α*S* and *1R-trans-*α*S* enantiomers. Given the enantioselective toxicity, it may be expected that the potential ecotoxicological effects of CP and analogues (e.g., cyfluthrin) will depend closely on the environmental behavior of the *1R-cis-*α*S** and *1R-trans-*α*S** enantiomers.

Isomer Selectivity in Degradation by Bacteria Isolates. Three strains of SP-degrading bacteria, *Vibrio hollisae* (CF-3), *Burkhoderia picketti* (CF-17), and *Erwinia carotovora* (CF-28), were isolated from sediment and used to degrade CP in solution media. Dissipation of CP diastereomers and cis enantiomers was

Table 1. Relative Contents of Diastereomers and Enantiomers and Measured LC50 Values (μg L^{-1}) for *C. dubia* for Various Cypermethrin (CP) Formulations

	relative content (%)							
	cis				trans		active content[b]	LC_{50}
formulation	*1R-cis-αR*	*1S-cis-αS*	*1R-cis-αS*[a]	*1S-cis-αR*	*1R-trans-αR* + *1S-trans-αR*	*1S-trans-αR* + *1R-trans-αS*[a]	(%)	(μg L^{-1})
CP	14.5	15.6	12.3	12.2	26.0	19.4	22.0	0.889
α-CP	8.6	8.9	41.4	41.1	0	0	41.4	0.696
β-CP	2.9	2.8	14.4	15.0	11.4	53.6	41.2	0.690
θ-CP	17.0	0	83.0	0	0	0	83.0	0.356

[a] Denotes insecticidally active enantiomers. [b] Content of *1R-cis-αS** and *1R-trans-αS** in the entire formulation.

Table 2. First-Order Rate Constant k (h^{-1}), Half-Life $T_{1/2}$ (h), and Correlation Coefficient r^2 for the Degradation of Cypermethrin Diastereomers by Three Synthetic Pyrethroid Degrading Bacteria as Determined by Achiral GC Analysis

		cis		trans	
bacteria		*1R-cis-αR* + *1S-cis-αS*	*1R-cis-αS*[a] + *1S-cis-αR*	*1S-trans-αS* + *1R-trans-αR*	*1S-trans-αR* + *1R-trans-αS*[a]
CF-3	k	0.0170	0.0116	0.0242	0.0231
	$T_{1/2}$	40.8	57.7	28.6	30.0
	r^2	0.99	0.94	0.96	0.97
CF-17	k	0.0207	0.0198	0.0318	0.0296
	$T_{1/2}$	33.5	35.0	21.8	23.4
	r^2	0.92	0.91	0.92	0.92
CF-28	k	0.0135	0.0135	0.0337	0.0308
	$T_{1/2}$	51.3	51.3	20.6	22.5
	r^2	0.93	0.92	0.93	0.94

[a] Denotes insecticidally active enantiomers.

Table 3. First-Order Rate Constant k (h^{-1}), Half-Life $T_{1/2}$ (h), and Correlation Coefficient r^2 for the Degradation of Cypermethrin Enantiomers by Three Synthetic Pyrethroid Degrading Bacteria as Determined by Chiral GC Analysis

		cis				trans	
bacteria		*1R-cis-αR*	*1S-cis-αS*	*1R-cis-αS*[a]	*1S-cis-αR*	*1S-trans-αS* + *1R-trans-αR*	*1S-trans-αR* + *1R-trans-αS*[a]
CF-3	k	0.0175	0.0206	0.0172	0.0196	0.0303	0.0412
	$T_{1/2}$	39.6	33.6	40.3	35.4	22.9	16.8
	r^2	0.97	0.96	0.95	0.96	0.94	0.98
CF-17	K	0.0201	0.0223	0.0206	0.0236	0.0318	0.0366
	$T_{1/2}$	34.5	31.1	33.6	29.3	21.8	18.9
	r^2	0.88	0.87	0.87	0.88	0.97	0.91
CF-28	k	0.0184	0.0221	0.0186	0.0217	0.0423	0.0443
	$T_{1/2}$	37.7	31.4	37.3	31.9	16.4	15.6
	r^2	0.93	0.91	0.92	0.91	0.96	0.95

[a] Denotes insecticidally active enantiomers.

fitted to a first-order decay model to estimate the rate constant k (h^{-1}) and half-life $T_{1/2}$ (h) (**Tables 2** and **3**). The fit was generally good, with $r^2 \geq 0.91$ for the diastereomers (**Table 2**) and $r^2 \geq 0.87$ for the resolved enantiomers (**Table 3**).

Degradation of CP diastereomers by the bacteria isolates was relatively rapid, and the longest $T_{1/2}$ was ~58 h. In the noninoculated solution, CP was found to be relatively stable, with the overall loss <10% after 112 h of incubation (data not shown). In the inoculated treatments, differences existed among the different bacteria isolates in their potential to transform the same diastereomer. More rapid degradation of the cis diastereomers occurred with CF-17, whereas faster degradation of the trans diastereomers was observed with CF-17 and CF-28 (**Table 2**). For the same degrader, the trans diastereomers were consistently degraded preferentially over the cis diastereomers, as shown for CF-28 in **Figure 4**. The selectivity in diastereomer degradation resulted in relative enrichment of the cis diastereomers, as shown in **Figure 2b** for CF-3. The isomer selectivity may be quantitatively characterized by diastereomer ratio DR, which was calculated as the ratio of the k values for the trans diastereomers over those for the cis diastereomers, or

$$\mathrm{DR} = \frac{k_{trans-1} + k_{trans-2}}{k_{cis-1} + k_{cis-2}} \quad (1)$$

where the subscripts *cis*-1, *cis*-2, *trans*-1, and *trans*-2 denote the diastereomers listed in the order given in **Table 2**. When DR is greater than 1.0, the trans diastereomers are degraded preferentially over the cis diastereomers. The estimated DR increased in the order CF-17 (1.52) < CF-3 (1.65) < CF-28 (2.39). This suggests that the trans diastereomers in CP were selectively degraded by all degraders and that CF-28 displayed greater selectivity for the trans diastereomers than the other two degraders. Using ^{14}C-labeled isomers, Sakata et al. (*17*) measured degradation of CP stereoisomers in two soils and observed significantly faster degradation for the trans diastereomers than for the cis diastereomers. Using the listed $T_{1/2}$ values, DR was estimated to be 3.59 for a light clay soil and

Table 4. First-Order Rate Constant k (d^{-1}), Half-Life $T_{1/2}$ (d), and Correlation Coefficient r^2 for the Degradation of Cypermethrin Diastereomers (Determined by Achiral GC Analysis) and Enantiomers (Determined by Chiral GC Analysis) in Sediment

	Diastereomers			
	cis		trans	
	1R-cis-αR + 1S-cis-αS	*1R-cis-αS*[a] *+ 1S-cis-αR*	*1S-trans-αS + 1R-trans-αR*	*1S-trans-αR + 1R-trans-αS*[a]
k	0.0122	0.0100	0.0175	0.0208
$T_{1/2}$	56.8	69.3	39.6	33.3
r^2	0.98	0.99	0.99	0.99

	Enantiomers					
	cis				trans	
	1R-cis-αR	*1S-cis-αS*	*1R-cis-αS*[a]	*1S-cis-αR*	*1S-trans-αS + 1R-trans-αR*	*1S-trans-αR + 1R-trans-αS*[a]
k	0.0088	0.0173	0.0093	0.0158	0.0220	0.0171
$T_{1/2}$	78.7	40.1	74.5	43.8	31.5	40.5
r^2	0.94	0.99	0.99	0.99	0.95	0.93

[a] Denotes insecticidally active enantiomers.

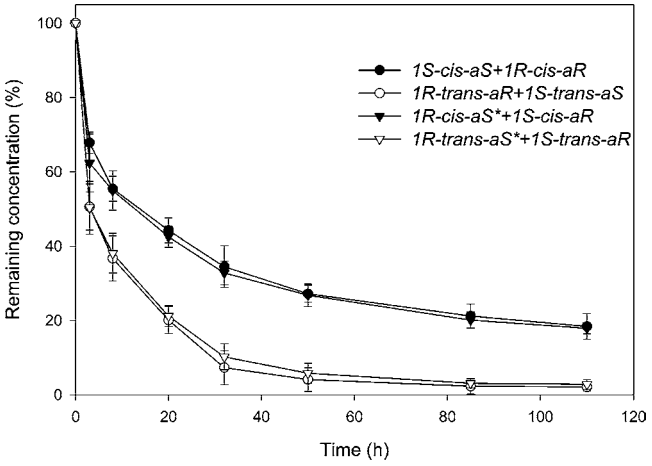

Figure 4. Degradation of cypermethrin diastereomers by a bacteria isolate CF-28 (*Erwinia carotovora*) at 21 °C in an inoculated mineral salt medium. (* denotes biologically active enantiomers).

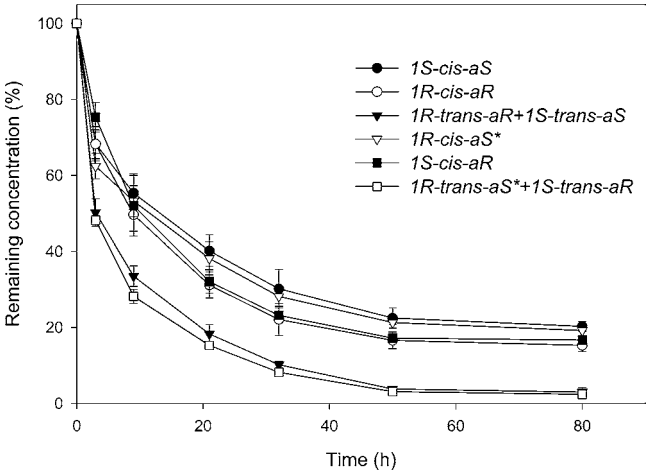

Figure 5. Degradation of cypermethrin enantiomers or diastereomers (enantiomer pairs) by a bacteria isolate CF −28 (*Erwinia carotovora*) at 21 °C in an inoculated mineral salt medium. (* denotes biologically active enantiomers).

3.08 for a sandy clay loam soil. Isomer selectivity was also observed for other SP compounds. For instance, the trans isomer of permethrin was found to degrade faster than the cis isomer in soils in two studies (*17*, *21*). In sediment and runoff water samples collected at a nursery site, *trans*-permethrin was consistently detected at lower concentrations than *cis*-permethrin (*7*).

The incomplete separation of enantiomers in the two trans diastereomers prevented estimation of k or $T_{1/2}$ for trans enantiomers. Enantiomers from the cis diastereomers were degraded more slowly by all bacteria strains than the unresolved trans diastereomers, as shown for CF-28 in **Figure 5**. The enantioselectivity resulted in relative enrichment of cis enantiomers, as shown for CF-28 in **Figure 3b**. The $T_{1/2}$ of *1R-cis-αS** was relatively long compared to the other stereoisomers for the same degrading bacteria (**Table 3**). The degradation kinetics for the other active enantiomer, *1R-trans-αS**, was unavailable from the data due to the lack of separation for the trans diastereomers. However, the unresolved enantiomer pair (*1S-trans-αR + 1R-trans-αS**) containing the active enantiomer appeared to degrade faster than the other diastereomers and resolved enantiomers (**Figure 5**, **Table 3**). In the Sakata et al. soil study, *1R-trans-αS** of CP was found to degrade substantially faster than the corresponding *1S-trans-αR*, and the ratio of the k values (*1R-trans-αS** over *1S-trans-αR*) was estimated to be 2.11 for a light clay soil and 1.80 for a sandy loam soil (*17*). Assuming similar selectivity for CP biodegradation in this study, *1R-trans-αS** may be expected to be the least persistent among all CP stereoisomers. Therefore, the difference between *1R-cis-αS** and *1R-trans-αS** in persistence may be compensatory and the overall persistence of the biologically active enantiomers may be similar to the overall trend of all CP stereoisomers. However, studies on other chiral compounds showed that enantioselectivity may change in response to environmental conditions. For instance, 1′*S*-(+) metalaxyl was selectively degraded in sewage sludge, but 1′*R*-(−) metalaxyl was preferentially degraded in soil (*22*). In another study, although 1′*R*-(−) metalaxyl was degraded faster than 1′*S*-(+) metalaxyl in a German soil, the opposite occurred in a Cameroonian soil (*23*). Environmental conditions were also found to dictate enantioselectivity in degradation of the herbicide dichlorprop in soils (*13*). Further method development is needed to separate the trans enantiomers in CP, and the selectivity between biologically active and inactive enantiomers in CP should be evaluated in other environments.

Isomer Selectivity in Degradation in Whole Sediment. Isomer selectivity in CP degradation was further evaluated in whole sediment. The dissipation of CP diastereomers or resolved enantiomers was fitted to a first-order kinetics, and the fit was good, with $r^2 \geq 0.98$ for the diastereomers and $r^2 \geq 0.93$ for the enantiomers (**Table 4**). Among the diastereomers, both cis

diastereomers were considerably more persistent than the trans diastereomers in the sediment, which was consistent with the results from the biodegradation experiment. The selectivity measured as DR was calculated to be 1.73. The $T_{1/2}$ for *1R-cis-*α*S** (74.5 d) was among the longest, whereas that of *1R-trans-*α*S** was probably among the shortest. The overall agreement between bacteria isolates and the whole sediment suggests that the isomer selectivity in CP degradation in sediment was biologically mediated and that isomer selectivity may occur widely for CP or similar SP compounds in the environment.

In conclusion, synthetic pyrethroids such as CP are made of multiple stereoisomers that differ greatly in biological activity. This study showed that only two (*1R-cis-*α*S** and *1R-trans-*α*S**) of the eight enantiomers are toxic to *C. dubia*, and the isomer selectivity in the measured aquatic toxicity was similar to that in insecticidal activity. Selected bacteria preferentially degraded some diastereomers or enantiomers over the others in both solution media and in sediment. Compared with the cis diastereomers, degradation of trans diastereomers was consistently more rapid, which resulted in relative enrichment of the cis diastereomers. Of the two biologically active enantiomers, *1R-cis-*α*S** was relatively persistent compared with the other stereoisomers, whereas *1R-trans-*α*S** was likely the least persistent. The selectivity in biodegradation may have great implications as ecotoxicological effects may largely be determined by the behavior of the biologically active enantiomers. Such selectivity should be considered in assessment of environmental risks of CP and analogues. Isomer selectivity may be compound and environment-specific and should be further determined for CP under other environmental conditions and for other SP compounds in both terrestrial and aquatic ecosystems.

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