Environ. Sci. Technol. **1998**, *32,* 2070−2076

Changes in the Enantiomeric Ratio of (*R*)- to (*S*)-Mecoprop Indicate in Situ Biodegradation of This Chiral Herbicide in a Polluted Aquifer

CHRISTIAN ZIPPER,[†] MARC J.-F. SUTER,
STEFAN B. HADERLEIN,
MICHAEL GRUHL,[‡] AND
HANS-PETER E. KOHLER*

Swiss Federal Institute for Environmental Sciences and Technology (EAWAG), Swiss Federal Institute of Technology (ETH), CH-8600 Dübendorf, Switzerland, CSD Colombi Schmutz Dorthe AG, CH-5000 Aarau, Switzerland

Leachate samples from a waste disposal site in Switzerland and groundwater samples downstream of the landfill were analyzed for residues of (*R*)- and (*S*)-mecoprop [(*R*)- and (*S*)-2-(4-chloro-2-methylphenoxy)propionic acid] by means of enantiomer-specific gas chromatography combined with mass spectrometry. (*R*)- And (*S*)-mecoprop were found at equal concentrations (up to 124 μg/L) in the landfill leachate, indicating that a racemic mixture of mecoprop leached to the receiving groundwater. Groundwater samples downstream of the landfill contained from <0.001 to 975 μg/L of mecoprop and 16 out of 31 samples showed a significant excess of (*R*)-mecoprop. Sorption of (*R*)- and (*S*)-mecoprop to aquifer matrix and to various reference minerals was generally low and did not discriminate between the mecoprop enantiomers. We conclude that enantioselective microbial degradation increased the enantiomeric ratio of (*R*)- to (*S*)-mecoprop during groundwater passage of the landfill leachate. Most leachate-affected groundwaters are subject to a very complex input pattern of contaminants in time and space. Thus, observations of concentration changes of contaminants with time or space are rarely indicative of specific transformation processes in such aquifers. Under such conditions, analysis of the enantiomeric ratio of chiral contaminants such as mecoprop can provide information about in situ biodegradation processes.

Introduction

Numerous xenobiotic organic compounds are typically present in landfill leachates (*1*−*3*). Recently, phenoxyalkanoic acid herbicides (*3*) and especially mecoprop were identified in leachates of many landfills (*4*−*8*). Although these compounds may account for only a few percent of the total organic matter present in the leachate, they are of considerable concern due to potential health risks. The water quality of groundwaters may strongly be affected by landfill leachates unless the toxic compounds are substantially attenuated in the leachate plumes. Attenuation of such compounds is primarily due to dilution, sorption, chemical reaction, and microbial degradation (*1*, *3*).

In a complex groundwater regime, it is often difficult to determine which of these processes governs the attenuation of a particular organic compound. For chiral organic compounds, a very promising way to assess the significance of microbial degradation among these processes within a leachate plume is to monitor the enantiomeric ratio. In general, microorganisms tend to enantioselectively degrade chiral organic compounds (*9*−*11*), e.g., (*S*)-mecoprop was preferentially degraded under aerobic conditions in soil (*12*), in lake water samples (*13*), in sewage sludge (*14*), and by *Sphingomonas herbicidovorans* (*15*). Reversed enantioselectivity was found in lake water samples (*13*) and in degradation experiments with *Alcaligenes denitrificans* (*16*). Different enantiomeric selectivity in different processes was also observed for dichlorprop. For soil, preferential degradation of (*S*)-dichlorprop was reported (*17*), whereas marine microorganisms preferentially degraded (*R*)-dichlorprop. In all of the above examples, it was shown that biological processes were responsible for the preferential degradation of one of the enantiomers. Therefore, changes in the enantiomeric ratio of a chiral organic compound in environmental samples indicate that the compound has been subject to microbial degradation, because other attenuation processes, such as dilution, diffusion, transport, and chemical reactions in an unsymmetrical environment (*18*), are known to be not enantioselective.

This paper describes a field study of chiral mecoprop [(*RS*)-2-(4-chloro-2-methylphenoxy)propionic acid (Figure 1, panels a and b)] in a contaminated aquifer downstream from a former waste disposal site in Switzerland. We provided evidence for enantioselective in situ biodegradation of mecoprop in the leachate-contaminated aquifer and ruled out other potentially enantioselective processes such as sorption to the aquifer matrix. Our results are based on an enantiomer-specific gas chromatography−mass spectrometry (GC/MS) method that allowed the detection of very low concentrations of (*R*)- and (*S*)-mecoprop in water samples with a complex background matrix such as a landfill leachate.

Experimental Section

The Waste Disposal Site. The waste disposal site Kölliken (Canton Aargau, Switzerland) is a former quarry for clays. It is situated in the aquitanian molasse, which is composed primarily of fractured marls and interlayered with banks of calcareous sandstone (*19*). Groundwater temperatures downstream of the disposal site are between 11.0 and 15.6 °C. The water table at a depth of about 3 m below surface exhibits a hydraulic gradient of 25−30 ‰ from north to south and seasonal fluctuations of up to 1 m. A mean hydraulic conductivity of 3.8×10^{-6} m/s was derived from the breakthrough of chloride and bromide from the wastes and from data obtained by hydraulic tests applying Darcy's law. From 1979 to 1985 chemical and partly toxic wastes were deposited in the landfill. The total waste volume of 300 000 m^3 consists of production residues from the chemical industry (chlorobenzenes, chlorophenols, and anilines), soil from excavation of contaminated sites (oil and various chemicals), domestic wastes, incinerator slug, and slug from an aluminum melting plant containing high amounts of salts. Furthermore, pesticides of an unknown composition and quantity were disposed of at the site. Leachate from the landfill originates from eight sectors, designated as S1−S8 (see Figure 2), and is collected by an efficient drainage system, which allows individual sampling and monitoring. Only sector S8 cannot

---

* Corresponding author: Department of Microbiology, EAWAG, Ueberlandstrasse 133, CH-8600 Duebendorf, Switzerland. Phone: +41 1 823 5521; fax: +41 1 823 5547; e-mail: kohler@eawag.ch.

[†] Present address: BMG Engineering, Ifangstrasse 11, CH-8952 Schlieren, Switzerland.

[‡] CSD Colombi Schmutz Dorthe AG. Present address: SMDK, Safenwilerstrasse 27, CH-5742 Kölliken, Switzerland.

S0013-936X(97)00880-8 CCC: $15.00

© 1998 American Chemical Society
Published on Web 06/05/1998

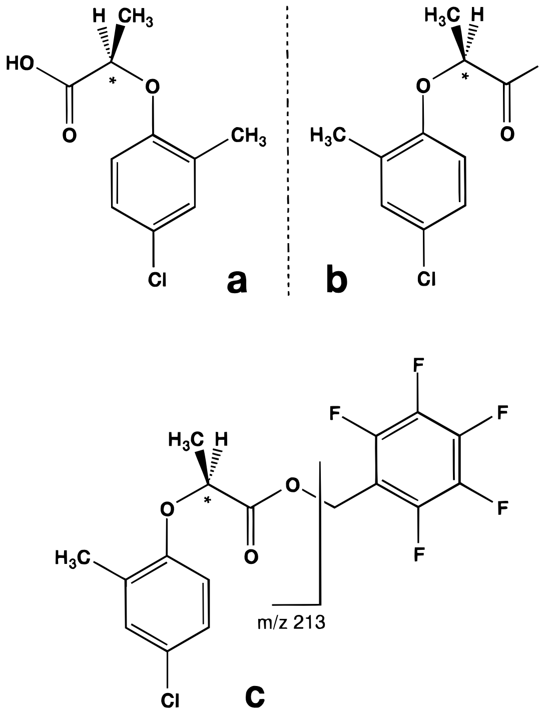

FIGURE 1. Structural formulas of (a) (*R*)-2-(4-chloro-2-methylphenoxy)propionic acid [(*R*)-mecoprop], (b) (*S*)-2-(4-chloro-2-methylphenoxy)propionic acid [(*S*)-mecoprop], and (c) of the corresponding 2,3,4,5,6-pentafluorobenzyl ester of (*S*)-mecoprop. The fragment ion *m/z* 213 was monitored under negative chemical ionization conditions.

be sampled individually, but information about the water pollution in leachate water of this sector can be taken from samples of well P0, which is a joint leachate water well of sectors S2, S3, S4, and S8. Since 1979, most of the landfill leachate and almost all of the landfill gas is collected and physically, chemically, and biologically treated. The amount of collected landfill leachate depends on precipitation and normally ranges from 50 to 150 m^3/day, but a small amount of highly contaminated water still leaches to the underlying groundwater (approximately 3 m^3/day). For monitoring the groundwater quality, over 150 groundwater wells (designated as KB) were drilled and water samples are analyzed in regular time intervals. The depth of the monitored section is designated by a suffix A, B, C, or D, where A represents the shallowest and D the deepest section of a particular well. The groundwater upstream of the disposal site was unpolluted and showed aerobic to nitrate-reducing conditions, whereas groundwater samples downstream and close to the disposal site showed sulfidogenic to methanogenic conditions. The complex local groundwater regime did not allow quantitative interpretations of analytical data, with one exception. It is known from the breakthrough of chloride and bromide (data not shown) that leachate from landfill sector S6 flows to the groundwater well KB37B. Untreated landfill leachate was generally heavily contaminated and contained an average of up to 6 mg/L of aniline and various chloroanilines, up to 1.3 mg/L of phenol and various chlorophenols, up to 40 μg/L of benzene, toluene, chlorobenzene, dichloromethane, up to 25 g/L chloride, 1−3 g/L of sulfate, 10−30 mg/L of nitrate, and 100−500 mg/L of dissolved organic carbon.

Storage, Extraction, and Derivatization of Samples. Leachate and groundwater samples (approximately 600 mL) were stored up to 7 months in 1 L glass flasks at −20 °C. For extraction, the samples were thawed and allowed to reach ambient temperature. Appropriate volumes (1−100 mL) were transferred to 100 or 250 mL glass Schott flasks and spiked with 25 μL of the spike standard solution [0.15 μg (*RS*)-dichlorprop-*ring*-$^{13}C_6$]. Each sample was then acidified to pH 1 with H_2SO_4, 10 mL of dichloromethane was added, and the extraction mixture was vigorously shaken. After setting aside the organic phase, the water phase was reextracted twice with 5 and 3 mL of dichloromethane. The combined extracts were centrifuged (10 min; 4000*g*), transferred to clean glass vials, and concentrated to approximately 10 mL under a gentle flow of nitrogen at 30 °C. For derivatization (*20−25*), 2 mL of acetone and 2 drops of K_2CO_3 solution (30% w/v in H_2O) were added and the solution was evaporated to approximately 1 mL. To remove the dichloromethane, 5 mL of acetone was added and evaporation to 1 mL was repeated twice. Then, 1 mL of acetone and 200 μL of 2,3,4,5,6-pentafluorobenzylbromide (PFBBr) solution (1% vol/vol in acetone) were added, and the solution was vigorously shaken and left overnight at room temperature for esterificiation. Then, 2 mL of *n*-hexane and 4 mL of H_2O were added to the samples, the vials were vigorously shaken, and the hexane phases were transferred to clean vials. Approximately 5 mg of anhydrous sodium sulfate was added, the solution filtered through a 0.5 μm filter (Millex LCR4, Millipore Corp.), and again transferred to clean vials. Standard solutions of the analyte and the spike standard were derivatized accordingly. Samples and standards were stored in Teflon-stoppered vials in the dark at 4 °C until analyzed.

Enantiomer-Selective GC−MS Analysis of Mecoprop. 2,3,4,5,6-Pentafluorobenzyl (PFB) esterification was considered to be the best method for mecoprop analysis in heavily contaminated landfill leachate samples when compared to other methods of derivatization, such as methylation or esterification with dicyclohexylcarbodiimide-2-chloroethanol. The use of a GC with electron capture detection [ECD sensitivities up to 4500-fold greater than the ones of the corresponding 2-chloroethyl esters (*21*)] during methods development and the excellent applicability for negative chemical ionization-mass spectrometry (NCI-MS) favored the highly fluorinated PFB derivative. In addition, the relatively long retention times of the PFB esters of phenoxyalkanoic acid herbicides allowed for better separation of the analytes from interfering contaminants. A GC 8065 (Fisons Instruments, Manchester, U.K.) coupled with a Micromass AutoSpec-Q double focusing magnetic sector mass spectrometer (Micromass, Manchester, U.K.) was used. The PFB esters of (*R*)- and (*S*)-mecoprop (Figure 1c) were separated on a 15 m glass column (0.25 mm i.d.) with an OV1701 polysiloxane phase containing 35% heptakis(2,3-dimethyl-6-*tert*-butyldimethylsilyl)-β-cyclodextrin (TBDM-β-CD) as the chiral selector (*12*). They were nearly baseline separated on this column (enantiomer resolution $R = 0.9$, Figure 3). Samples (1 μL) were injected on-column at 60 °C, and the oven temperature was programmed as follows: 2 min at 60 °C, 20 °C/min to 186 °C, 2 °C/min to 196 °C, then 25 °C/min to 230 °C followed by an isothermal hold for 1 min. All samples were analyzed by negative chemical ionization (NCI, CI gas: isobutane, 40 eV) with selected ion monitoring (SIM). Two ions, *m*/*z* 213.0318 and *m*/*z* 238.9974, were monitored simultaneously (0.5 s/channel) for the detection of (*R*)- and (*S*)-mecoprop and (*R*)- and (*S*)-dichlorprop-*ring*-$^{13}C_6$, respectively, using a lockmass of *m*/*z* 230.9856 from perfluorokerosene. Concentrations of (*R*)- and (*S*)-mecoprop in the samples were determined by external calibration and corrected for sample size and recovery. The external standard had an (*RS*)-mecoprop concentration of 0.2565 μg/mL in *n*-hexane, MS response being linear up to this concentration. Samples at higher concentrations were diluted. Recoveries were determined from peak area ratios relative to the spike standard (*RS*)-dichlorprop-*ring*-$^{13}C_6$. Replicate determina-

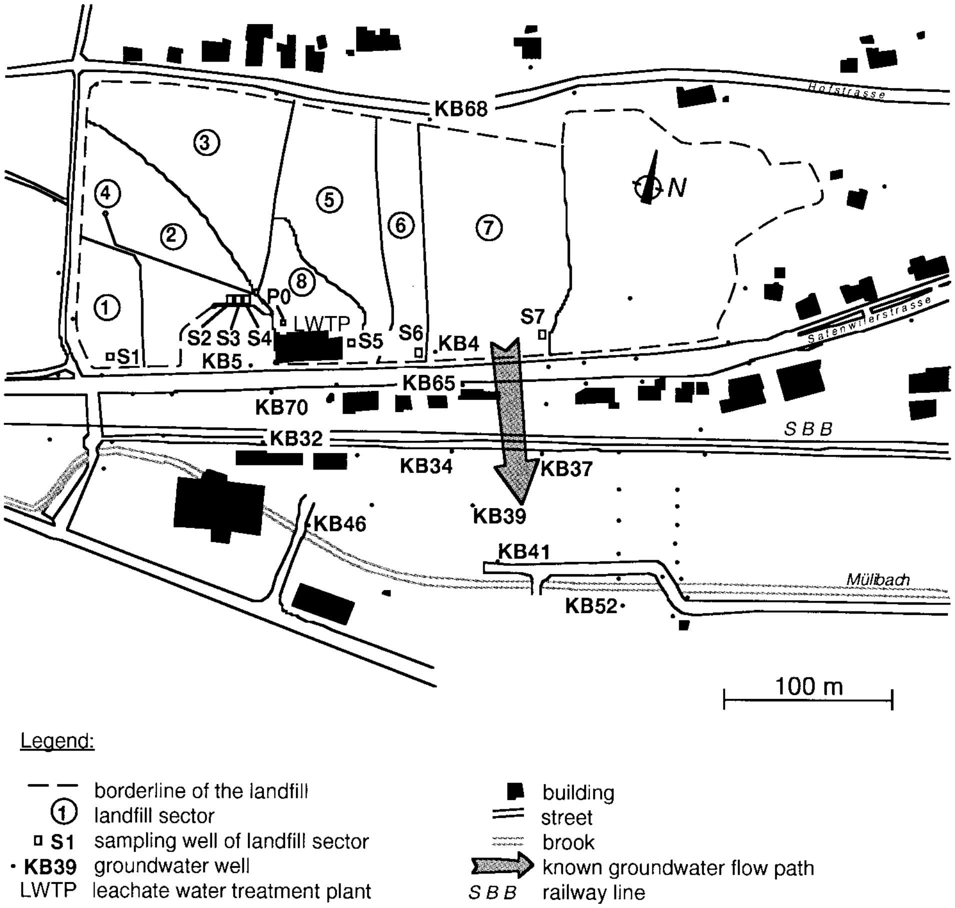

FIGURE 2. Map of the waste disposal site Kölliken, Canton Aargau, Switzerland.

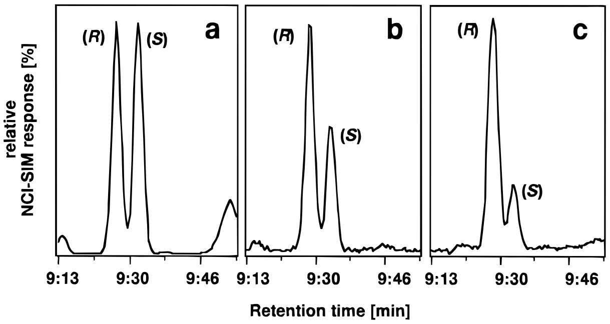

FIGURE 3. NCI-SIM chromatograms (*m/z* 213.0318) of (*R*)- and (*S*)-mecoprop. Panel a shows a sample of the landfill leachate of sector S2 taken February 15, 1996, and panels b and c show two groundwater samples downstream of the landfill taken at the wells (b) KB34C on February 15, 1996, and (c) KB52A on November 15, 1995. The ER of (*R*)- to (*S*)-mecoprop was 1.00 (a), 1.68 (b), and 4.04 (c), respectively.

tions ($n \geq 2$) were made for selected samples. Some samples were reanalyzed using full-scan NCI high-resolution mass spectrometry ($m/z = 50-400$, 1 s/decade, resolution $M/\Delta M = 12\,000$) for confirmation. The recoveries obtained with the PFB derivatives in heavily contaminated landfill leachates were 78 ± 30% ($n = 64$) for the spike standard, which is only slightly lower than values reported for unpolluted surface waters (91 ± 4%) (*25*). Average RSD values of the concentration measurements of 14.5 and 14.7% ($n = 20$) for (*R*)- and (*S*)-mecoprop, respectively, were calculated for leachate and groundwater samples. The detection limit of 0.001 μg/L for (*R*)- and (*S*)-mecoprop in heavily contaminated landfill leachate was 1 order of magnitude lower than that reported for rainwater (*26*) and 50−100 times lower than that published for surface waters (*25*).

TABLE 1. Concentrations[a] of (*R*)- and (*S*)-Mecoprop and Enantiomeric Ratios[a] in Landfill Leachates of the Waste Disposal Site Kölliken, Taken on February 15, 1996

landfill leachate from sector	(*R*)-mecoprop ± sd (μg/L)	(*S*)-mecoprop ± sd (μg/L)	ER[b] ± sd
S1	0.077 ± 0.001	0.075 ± 0.001	1.02 ± 0.01
S2	14.0 ± 0.8	14.0 ± 1.1	1.00 ± 0.01
S3	46.9 ± 0.2	46.4 ± 0.1	1.01 ± 0.01
S4	6.4 ± 1.2	6.6 ± 1.3	0.98 ± 0.03
S5	18.5 ± 0.1	18.5 ± 0.1	1.00 ± 0.01
S6	64.9 ± 4.8	58.6 ± 3.9	**1.11** ± 0.03
S7	2.5 ± 0.1	2.4 ± 0.1	1.02 ± 0.02
P0[c]	26.9 ± 0.3	27.1 ± 0.6	1.00 ± 0.01

[a] Values represent means ± standard deviations of at least two independent samples. [b] The enantiomeric ratio was defined as the peak area of (*R*)-mecoprop divided by the peak area of (*S*)-mecoprop. Significant values are printed bold. [c] Joint sample of landfill sectors S2, S3, S4, and S8.

Enantiomeric ratios (ER) were defined as the peak area of (*R*)-mecoprop divided by the peak area of (*S*)-mecoprop, assuming identical MS response for both enantiomers. The enantiomer resolution of the PFB ester derivatives made possible the determination of a wide range of ER values ($0.05 \leq \mathrm{ER} \leq 20$). The precision of the determination of ER values in heavily contaminated samples, as well as in standards, was excellent with an RSD of 3.0% ($n = 20$). On the basis of these results, we concluded that alterations of the ER of more than three times the RSD were significant.

Sorption Experiments. Sorption experiments were carried out with aquifer matrix and model mineral surfaces as sorbents (for specifications see Table 3). Unpolluted and representative samples of marl and sandstone, the dominant

TABLE 2. Concentrations[a] of (*R*)- and (*S*)-Mecoprop and Enantiomeric Ratios[a] in Groundwater from the Leachate Plume of the Waste Disposal Site Kölliken, Taken on November 15, 1995, and February 15, 1996

		sampling date: November 15, 1995			sampling date: February 15, 1996		
observation well	depth of sampling (m)	(*R*)-mecoprop ± sd (μg/L)	(*S*)-mecoprop ± sd (μg/L)	ER[b] ± sd	(*R*)-mecoprop ± sd (μg/L)	(*S*)-mecoprop ± sd (μg/L)	ER[b] ± sd
KB 4	7	0.011 ± 0.002	0.011 ± 0.002	0.99 ± 0.01	0.013 ± 0.003	0.013 ± 0.003	0.98 ± 0.01
KB 5	5.5	0.025	0.027	0.91	0.002	0.002	1.00
KB 32A	9	0.016	0.013	**1.21**	0.010	0.009	**1.10**
KB 34B	15	1.51	1.61	0.93	14.18	11.95	**1.10**
KB 34C	20	64.6 ± 12.1	64.9 ± 12.1	1.00 ± 0.02	0.322 ± 0.007	0.194 ± 0.028	**1.68** ± 0.25
KB 37B	18	1.38 ± 0.36	1.06 ± 0.23	**1.31** ± 0.06	4.51 ± 0.37	3.50 ± 0.29	**1.28** ± 0.04
KB 37C	25	13.5 ± 5.5	14.6 ± 6.0	0.94 ± 0.02	9.1 ± 5.1	9.1 ± 4.5	0.97 ± 0.07
KB 39C	23	215 ± 51	208 ± 50	1.03 ± 0.00	488 ± 20	487 ± 19	1.00 ± 0.02
KB 41B	16	0.57	0.53	1.08	2.11	1.84	**1.13**
KB 41C	19	0.57	0.51	**1.12**	2.01	1.83	1.09
KB 41D	27	0.66 ± 0.34	0.62 ± 0.27	1.04 ± 0.08	2.29 ± 0.16	2.11 ± 0.07	1.08 ± 0.03
KB 46B	14	0.019	0.007	**2.80**	nd[c]	nd	nd
KB 46C	20	0.018	0.006	**2.81**	0.009	0.007	**1.25**
KB 46D	24	0.015	0.005	**3.28**	<0.001	<0.001	na[d]
KB 52A	7	0.012	0.003	**4.04**	<0.001	<0.001	na
KB 52B	15.5	0.013	0.004	**3.13**	<0.001	<0.001	na
KB 65A	33	0.012	0.002	**7.63**	0.041	0.042	0.98
KB 70A	26	0.017	0.008	**2.30**	<0.001	<0.001	na

[a] The values represent means ± standard deviations of at least two independent samples. [b] The enantiomeric ratio was defined as the peak area of (*R*)-mecoprop divided by the peak area of (*S*)-mecoprop. Significant values are printed bold. [c] nd, not determined. [d] na, not applicable.

TABLE 3. Adsorption Coefficients, K_d, of (*R*)- and (*S*)-Mecoprop for Aquifer Matrix of the Waste Disposal Site Kölliken, and for Some Model Mineral Surfaces[a]

				adsorption coefficent (K_d)[b] (L/kg) for the experiments with			
				(*R*)-mecoprop at		(*S*)-mecoprop at	
sorbent	specification, supplier	external surfacearea (*S*) (m^2/g)	ratios of sediment to water (r_{sw}) (mg/mL)	0.182 mg/L	1.82 mg/L	0.182 mg/L	1.82 mg/L
sandstone[c]		4	620	<1	<1	<1	<1
marl[c]		12	570	<1	<1	<1	<1
amorphous SiO_2	Aerosil 380; Degussa	380	23	34 ± 6	24 ± 6	30 ± 4	21 ± 2
$CaCO_3$	21061, Fluka	5	230	<1	<1	<1	<1
montmorillonite	K10; Fluka	270	300	35 ± 9	20 ± 3	36 ± 10	23 ± 4
δ-Al_2O_3	Alox C, Degussa	96	530	1 ± 1	<1	1 ± 1	<1
goethite	Bayferrox 910, Bayer	15	200	<1	<1	<1	<1
MnO_2	63553, Fluka	90	250	<1	<1	<1	<1

[a] Experimental conditions: pH = 6.8; 0.1 M KCl; $T = 25 \pm 1.5$ °C. [b] Mean values ($n = 3$); standard deviations given where appropriate. [c] Taken from the borehole KB68 (Figure 2). The material used was the fraction <63 μm.

types of aquifer material, were taken upstream of the landfill at a depth of 11.2 and 12.0 m, respectively, from core material of borehole KB68 (see Figure 2). The aquifer material was air-dried (4 days at room temperature), crushed mechanically, and then sieved through a 0.063 mm sieve. The marl and the sandstone contained 0.2 and 0.3% of organic carbon, respectively. Batch sorption experiments were conducted in triplicate in 1.8 mL borosilicate glass vials with aluminum foil liners and septum screw caps (Supelco SA, Gland, Switzerland). The aqueous phase contained 0.1 M KCl, and the temperature was maintained at 25 ± 1.5 °C. The pH of the suspensions was 6.8 and was controlled by addition of acid or base to the spiked solutions (no buffers). Two initial aqueous concentrations of (*R*)- and (*S*)-mecoprop were used for the sorption experiments (0.182 and 1.820 mg/L). Particle concentrations ranged from 20 to 600 g of solids L^{-1}, resulting in a decrease of the initial solution-phase concentration of the solutes by 0−80%. The suspensions were shaken on a rotary shaker at 30 rpm for 4 h, which was sufficient to reach an apparent sorption equilibrium. Phase separation was achieved by centrifugation at 12 000 rpm for 4 min. Assays containing spike solutions of (*RS*)-mecoprop but no sorbents were processed in the same way as the suspension samples and served as blanks. Sorbed concentrations were calculated from the difference between initial and equilibrium solution-phase concentrations. Concentrations of (*R*)- and (*S*)-mecoprop were determined by an enantiomer-specific HPLC method described previously (*15*).

Chemicals. Pure (99%) (*RS*)- and (*R*)-mecoprop were obtained from Riedel-de Haen (Seelze, Germany). (*RS*)-Dichlorprop-*ring*-$^{13}C_6$ (with a chemical purity of greater than 98% and an isotopic purity of 99%) was purchased from Cambridge Isotope Laboratories, Innerberg, Switzerland. Anhydrous sodium sulfate (Fluka, Buchs, Switzerland) and 2,3,4,5,6-pentafluorobenzylbromide (Aldrich, Steinheim, Germany) were reagent grade. Dichloromethane and *n*-hexane were pesticide residue grade (Burdick and Jackson, Fluka, Buchs, Switzerland). All other chemicals were of the highest purity commercially available and were obtained from Fluka, Buchs, Switzerland.

Results and Discussion

(*R*)- and (*S*)-Mecoprop Concentrations and ER Values in Landfill Leachate. Landfill leachate samples taken on February 15, 1996, from different sectors contained from 0.077 to 64.9 μg/L (*R*)-mecoprop and from 0.075 to 58.6 μg/L (*S*)-mecoprop (Table 1). Mecoprop in all but one (S6, ER = 1.1) landfill sectors was racemic (Table 1). These data reflect the presence of mecoprop in all sectors of the landfill and an accumulation in sectors S3, S6, and probably in sector S8 (from the analysis of the sample taken from well P0). Since there is no information available about the formulation of the pesticides which were disposed of at this landfill, we cannot entirely exclude that wastes containing nonracemic mecoprop were dumped. However, because 94–98% of the landfill leachate is collected by the drainage system and, with only one exception, all ER values were close to 1, it is almost certain that racemic mecoprop leached to the receiving groundwater.

(*R*)- and (*S*)-Mecoprop Concentrations and ER Values in Groundwater from the Landfill Leachate Plume. Groundwater samples taken on November 16, 1995, downstream of the disposal site contained from 0.011 to 215 μg/L (*R*)-mecoprop and from 0.002 to 208 μg/L (*S*)-mecoprop (Table 2). Groundwater samples from the observation well KB39C contained (*R*)- and (*S*)-mecoprop at concentrations over 3 times higher than the highest concentrations measured in landfill leachate samples (Sector S6, Table 1). This indicates that the release of mecoprop in the landfill was not continuous but very variable, probably due to periodic breakage of mecoprop containers within the landfill. We also found high concentrations of (*R*)- and (*S*)-mecoprop in samples from the groundwater observation wells KB34C and KB37C (Table 2). All wells that contained high concentrations of mecoprop—KB34, KB37, and KB39—are located in the South-South-East direction from the disposal site and downstream of a known groundwater flow line (Figure 2). The ER values of 10 out of the 18 analyzed groundwater samples showed significant, up to a 7-fold, excess of (*R*)-mecoprop (Table 2). The highest ER (7.63) was found in well KB65A with 0.012 μg/L (*R*)-mecoprop and 0.002 μg/L (*S*)-mecoprop (Table 2). This well is located close to the disposal site (Figure 2) and is the deepest groundwater well sampled in this study ($d = 33$ m, Table 2). Well KB 70A is also located close to the disposal site in the deep groundwater zone and showed an ER of 2.3. All other wells with an excess of (*R*)-mecoprop—KB32A (ER = 1.21), KB37B (ER = 1.31), KB46B, C, and D (ER values of 2.80, 2.81, and 3.28, respectively), and KB52A and B (ER values of 4.04 and 3.13, respectively)—are situated rather far from the landfill (e.g., KB37, KB52) or are not located in the main groundwater flow path (e.g., KB32, KB46, see Figure 2).

Most groundwater samples taken on February 15, 1996, contained mecoprop concentrations comparable to those of the samples taken on November 15, 1995 (Table 2). In samples from the groundwater wells KB34C and KB37C, we found lower (*R*)- and (*S*)-mecoprop concentrations, whereas in KB39C, concentrations were higher than those measured on November 15, 1995 (Table 2). These data suggest that a water package with a high mecoprop concentration moved in the plume in a South-South-Easterly direction during the period between the first and the second sampling. This view is substantiated by the fact that higher mecoprop concentrations were found in the wells KB41B, C, and D. The ER values of 6 out of 13 groundwater wells showed a significant, up to 1.68-fold, excess of (*R*)-mecoprop. Some samples showed ER values (KB32A, KB37B) similar to those measured 3 months earlier, and in four samples—KB46D, KB52A, KB52B, and KB70A—neither the (*R*) nor the (*S*) enantiomer of mecoprop was detected anymore (Table 2).

Sorption of Mecoprop to Solid Surfaces. In the complex groundwater regime of the Kölliken site with non-steady-state leaching of mecoprop, enantioselective sorption of mecoprop to the aquifer matrix could contribute to the changes of the ER values observed in the groundwater downstream of the landfill. Different sorption properties would result in different fractions of the enantiomers in the aqueous phase and, thus, would cause different retardation and bioavailability of the compounds in the aquifer. To test this hypothesis, sorption of (*R*)- and (*S*)-mecoprop to aquifer matrix and to various minerals was studied at pH and ionic strength conditions similar to those prevailing in the aquifer (Table 3). The chosen pure mineral sorbents represent the predominant minerals and surface sites present in the aquifer. Carbonates and quartz make up the bulk of the aquifer matrix whereas iron and manganese (hydr)oxides, which tend to form coatings, as well as clay minerals, may better represent the prevailing surface sites of the matrix.

Generally, sorption of (*R*)- and (*S*)-mecoprop to the sorbents did not differ significantly (Table 3). Under the conditions studied, significant sorption occurred only with SiO_2 and montmorillonite, which have high specific surface areas. Sorption of both mecoprop enantiomers to aquifer matrix and to most of the model surfaces was too low to be quantified by our method (distribution coefficient, $K_d \leq 1$ L kg^{-1}). When normalized to the surface area, sorption of mecoprop to SiO_2 and montmorillonite was also quite low (K'_d on the order of 10^{-4} L m^{-2}). Taking this value as an upper limit for sorption to the aquifer matrix, the fraction of mecoprop present in the aqueous phase of the aquifer, f_{aq} (*27*)

$$f_{aq} = \frac{1}{1 + K'_d \rho_s S \frac{1-\epsilon}{\epsilon}} \qquad (1)$$

is calculated to be about 70% (porosity of the aquifer, $\epsilon = 0.35$; surface area of the aquifer matrix, $S = 1$ m^2 g^{-1}; density of the aquifer matrix, $\rho_s = 2500$ g L^{-1}). This corresponds to an average retardation factor R_f of about 1.4 [$R_f = (1/f_{aq})$].

The results suggest that enantioselective sorption was not responsible for the alterations in the ER values observed in groundwater samples from the leachate plume. Due to their low affinity for the aquifer matrix, bioavailability of both mecoprop stereoisomers was equally high and their retardation in the groundwater was very low and considered negligible for the purpose of this study. Our results are consistent with other studies reporting a high mobility of mecoprop and related organic anions in the subsurface (*28*–*30*).

Chemical Reactions and Dilution of Mecoprop During Groundwater Passage of the Landfill Leachate. Under environmental conditions, mecoprop is quite resistant to hydrolysis, reduction, and oxidation (*6*, *31*, *32*). Chemical reactions of mecoprop with organic or inorganic compounds present in the landfill leachate during groundwater passage cannot be completely ruled out, but they are expected to be very slow and not enantioselective. Abiotic enantioselective transformations require a chiral catalyst or an excess of one enantiomer of a reactive chiral compound (*18*), conditions which have not been reported for groundwater environments. Dilution of landfill leachate with infiltrating groundwater is not an enantioselective process and, therefore, cannot alter the ER.

Microbial Activity and Enantioselectivity in Microbial Degradation of Mecoprop During Groundwater Passage of the Landfill Leachate. The consumption of electron acceptors (oxygen, nitrate, sulfate) and dissolved organic carbon (DOC) and the production of dissolved reduced species [iron(II), sulfide, manganese(II), methane] indicate microbial activity in the leachate plume (e.g., refs *3* and *33*). Assuming steady state with respect to salt leaching (*19*), we estimated that 0.5 g/L sulfate was consumed during groundwater passage of landfill leachate from sector S6 to the well KB37B (Table 4). Analogous calculations revealed that also more than 13.5 mg/L of nitrate and 0.27 g/L DOC were consumed (Table 4). These estimates provide some evidence for denitrifying and sulfidogenic conditions between sector S6

TABLE 4. Measured and Expected Average Concentrations of the Conservative Tracer Chloride,[a] the Electron Acceptors Sulfate and Nitrate, and of Dissolved Organic Carbon (DOC) in Landfill Leachate Samples from Sector S6 and in Groundwater Samples from Well KB37B Downstream of the Landfill

compd or sum parameter	measured concn in landfill leachate from sector S6	measured concn in groundwater from well KB37B	expected concn in groundwater from well KB37B[b]	difference between measured and expected concn in KB37B
chloride (g/L)	22	15	15	0
sulfate (g/L)	3.0	1.6	2.1	−0.5
Nitrate (mg/L)	20	<0.5	14	>−13.5
DOC (g C/L)	0.4	0.01	0.28	−0.27

[a] A dilution factor of 0.3 was calculated from the chloride balance by comparing the concentrations measured in landfill leachate from sector S6 and the chloride concentrations determined in groundwater well KB37B. [b] Assuming a dilution factor of 0.3.

of the disposal site and the groundwater well KB37B. Since the input of mecoprop with landfill leachate was discontinuous and therefore the groundwater system was not at steady state with respect to mecoprop (see above), we cannot attribute the apparent depletion of mecoprop between sector S6 and well KB37B to degradation of mecoprop under denitrifying or sulfidogenic conditions. However, we can make sound conclusions regarding the processes that caused changes in the ER values.

So far, (*RS*)-mecoprop was studied in landfill leachates with analytical methods that could not differentiate between the two enantiomers. At the landfill of Vejen, Denmark, mecoprop was the most significant soluble organic contaminant in the plume but did not show any signs of degradation within the studied 130 m of the plume (*8*). Supplementary sampling showed that mecoprop disappeared further downstream. The authors could not determine whether mecoprop disappeared in the denitrifying or in the aerobic zone (*8*), but they stated that laboratory experiments have so far failed to show any mecoprop degradation under denitrifying conditions. In a model sulfidogenic aquifer, the three herbicides 2,4-dichlorophenoxyacetic acid, 2,4,5-trichlorophenoxyacetic acid, and 4-chloro-2-methylphenoxyacetic acid proved to be almost persistent. Only under aerobic conditions could biodegradation be observed after a lag period (*34*). These findings are interesting, because 2,4-dichlorophenoxyacetic and 2,4,5-trichlorophenoxyacetic acid are biotransformed under methanogenic conditions, but sulfate is able to inhibit the initial reaction (*35*). Under methanogenic conditions (incubations with sewage sludge), mecoprop seemed to be resistant to microbial attack and was not degraded after 49 days of incubation, whereas 2,4-D was completely removed after 17 days in the same experiment (*14*). These studies show that mecoprop is quite persistent under anaerobic field conditions.

However, mecoprop is known to degrade in aerobic environments by the action of microorganisms (*36*, *37*), and enantioselective microbial degradation was reported by various research groups (*12*, *14*, *15*, *38*). In most studies, preferential degradation of the (*S*)-mecoprop was found (*12*−*15*), but the findings of reversed enantioselectivity (*13*, *16*) suggest that probably each process has its own selectivity governed by specific enzymes. We observed ER values greater than 1 (Table 2), indicating that aerobic conditions were encountered during groundwater passage of the leachate.

Changes in the ER values do not give any information regarding absolute depletion of a chiral compound, but they are indicators for in situ biological transformations. Especially in complex groundwater environments—e.g., at landfills, such as the Kölliken site—data with regard to in situ biotransformation of organic pollutants is very valuable for assessing their long term fate in the aquifer and cannot be easily obtained otherwise.

Acknowledgments

We thank A. Klumpp, N. Astori, and R. Gloor (Institute Bachema AG, Schlieren, Switzerland) for providing water samples and for helping with the mecoprop analysis, M. D. Müller (Eidg. Forschungsanstalt, Wädenswil, Switzerland) for helping with the enantiomer separation of the PFB derivative of mecoprop, C. Martinson (CSD Colombi Schmutz Dorthe AG, Bern, Switzerland) for valuable information about the geochemistry and hydrology of the Kölliken landfill site, D. Kobler for determining surface areas, and J. L. Tardent (Sondermülldeponie Kölliken, Kölliken, Switzerland) for his support and interest in this study. We are grateful to K. Nickel, S. Resnick, E. Hoehn, and D. Zipper for critical reading of the manuscript.

Literature Cited

(1) Rügge, K.; Bjerg, P. L.; Christensen, T. H. *Environ. Sci. Technol.* **1995**, *29*, 1395−1400.
(2) Reinhard, M.; Goodman, N. L.; Barker, J. F. *Environ. Sci. Technol.* **1984**, *18*, 953−961.
(3) Christensen, T. H.; Kjeldsen, P.; Albrechtsen, H.-J.; Heron, G.; Nielsen, P. H.; Bjerg, P. L.; Holm, P. E. *Crit. Rev. Environ. Sci. Technol.* **1994**, *24*, 119−202.
(4) Kjeldsen, P. *J. Hydrol.* **1993**, *142*, 349−371.
(5) Öman, C.; Hynning, P. *Proceedings Sardinia 91, Third International Landfill Symposium*; CISA, Environmental Sanitary Engineering Centre: Cagliari, Italy, 1991; pp 857−864.
(6) Gintautas, P. A.; Daniel, S. R.; Macalady, D. L. *Environ. Sci. Technol.* **1992**, *26*, 517−521.
(7) Schultz, B.; Kjeldsen, P. *Water Res.* **1986**, *20*, 965−970.
(8) Lyngkilde, J.; Christensen, T. H. *J. Contamin. Hydrol.* **1992**, *10*, 291−307.
(9) Ariëns, E. J. In *Chiral Separations by HPLC*; Krstulovic, A. M., Ed.; Ellis Horwood Limited: Chichester, Great Britain, 1989; pp 31−68.
(10) Armstrong, D. W.; Reid, G. L.; Hilton, M. L.; Chang, C.-D. *Environ. Pollut.* **1993**, *79*, 51−58.
(11) Simoni, S.; Klinke, S.; Zipper, C.; Angst, W.; Kohler, H.-P. E. *Appl. Environ. Microbiol.* **1996**, *62*, 749−755.
(12) Müller, M. D.; Buser, H.-R. *Environ. Sci. Technol.* **1997**, *31*, 1953−1959.
(13) Buser, H.-R.; Müller, M. D. *Environ. Sci. Technol.* **1998**, *32*, 626−633.
(14) Zipper, C. Ph.D. Dissertation Thesis, Swiss Federal Institute of Technology (ETH), 1998.
(15) Zipper, C.; Nickel, K.; Angst, W.; Kohler, H.-P. E. *Appl. Environ. Microbiol.* **1996**, *62*, 4318−4322.
(16) Tett, V. A.; Willets, A. J.; Lappin-Scott, H. M. *FEMS Microbiol. Ecol.* **1994**, *14*, 191−200.
(17) Garrison, A. W.; Schmitt, P.; Martens, D.; Kettrup, A. *Environ. Sci. Technol.* **1996**, *30*, 2449−2455.
(18) March, J. *Advanced Organic Chemistry: Reactions, Mechanisms, and Structure*; John Wiley & Sons: New York, 1985.
(19) Martinson, C. *Eclogae geol. Helv.* **1994**, *87*, 473−486.
(20) Agemian, H.; Chau, A. S. Y. *Analyst* **1976**, *101*, 732−737.
(21) Agemian, H.; Chau, A. S. Y. *J. Assoc. Off. Anal. Chem.* **1977**, *60*, 1070−1076.
(22) Cotterill, E. G. *J. Chromatogr.* **1979**, *171*, 478−481.

(23) Chau, A. S. Y.; Terry, K. *J. Assoc. Off. Anal. Chem.* **1976**, *59*, 633−636.
(24) Lee, H.-B.; Stokker, Y. D.; Chau, A. S. Y. *J. Assoc. Off. Anal. Chem.* **1986**, *69*, 557−560.
(25) Vink, M.; van der Poll, J. M. *J. Chromatogr., A* **1996**, *733*, 361−366.
(26) Bucheli, T. D.; Grüebler, F. C.; Müller, S. R.; Schwarzenbach, R. P. *Anal. Chem.* **1997**, *69*, 1569−1576.
(27) Schwarzenbach, R. P.; Gschwend, P. M.; Imboden, D. M. *Environmental Organic Chemistry*; John Wiley & Sons: New York, 1993.
(28) Felding, G. *Sci. Total Environ.* **1995**, *168*, 11−18.
(29) Fielding, M.; Barcelo, D.; Helweg, A.; Galassi, S.; Torstensson, L.; Van Zoonen, P.; Wolter, R.; Angeletti, G. *Pesticides in Ground and Drinking Water*; Review Report 27, Commission of the European Communities, December 1991.
(30) Jafvert, C. T. *Environ. Toxicol. Chem.* **1990**, *9*, 1259−68.
(31) Worthing, C. R.; Hance, R. J. *The Pesticide Manual − A World Compendium*; The British Crop Protection Council: Farnham, Great Britain, 1991.
(32) Bovey, R. W.; Young, A. L. *The Science of 2,4,5-T and Associated Phenoxy Herbicides*; John Wiley & Sons: New York, 1980.
(33) Hunkeler, D.; Höhener, P.; Häner, A.; Bregnard, T.; Zeyer, J. In *Groundwater Quality, Remediation and Protection*; Kovas, K., Krasny, J., Eds.; IAHS: 1995; pp 421−430.
(34) Kuhlmann, B.; Kaczmarzcyk, B. *Environ. Toxicol. Water Qual.* **1995**, *10*, 119−125.
(35) Kuhn, E. P.; Suflita, J. M. In *Reactions and Movement of Organic Chemicals in Soil*; Sawhney, B. L., Brown, K., Eds.; Soils Science Society, Inc.: Madison, 1989.
(36) Smith, A. E.; Hayden, B. J. *Weed Res.* **1981**, *21*, 179−183.
(37) Smith, A. E.; Aubin, A. J. *J. Environ. Qual.* **1991**, *20*, 436−438.
(38) Heron, G.; Christensen, T. H. *Chemosphere* **1992**, *24*, 547−557.

Received for review October 7, 1997. Revised manuscript received April 13, 1998. Accepted April 21, 1998.

ES970880Q