Isomeric and Enantiomeric Composition of Different Commercial Toxaphenes and of Chlorination Products of (+)- and (−)-Camphenes

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The isomeric composition of commercial toxaphenes, including products of U.S. and former East German origin, and the enantiomeric composition of several key components were investigated using achiral and chiral high-resolution gas chromatography (HRGC) in combination with mass spectrometry (MS). Achiral HRGC with electron-capture, negative ionization (ECNI) MS was used for congener group analysis and chiral HRGC with electron ionization (EI) MS/MS for the isomer selective and enantioselective determination of some polychlorobornanes and polychlorobornenes. All products showed remarkably similar isomeric and enantiomeric compositions with some components present as almost racemic mixtures and others showing small but definite deviations from exact 1:1 enantiomeric ratios. Laboratory-scale chlorination of (+)- and (−)-camphene led to similar mixtures of polychlorobornanes with ≈50% of the enantiomeric excess retained. Chiroptical measurements showed a change in the direction of rotation of camphenes upon chlorination: the net rotation of the chlorination products of (−)-camphene was dextrorotatory, while those of (+)-camphene were levorotatory. The commercial toxaphenes showed a small net rotation that is levorotatory.

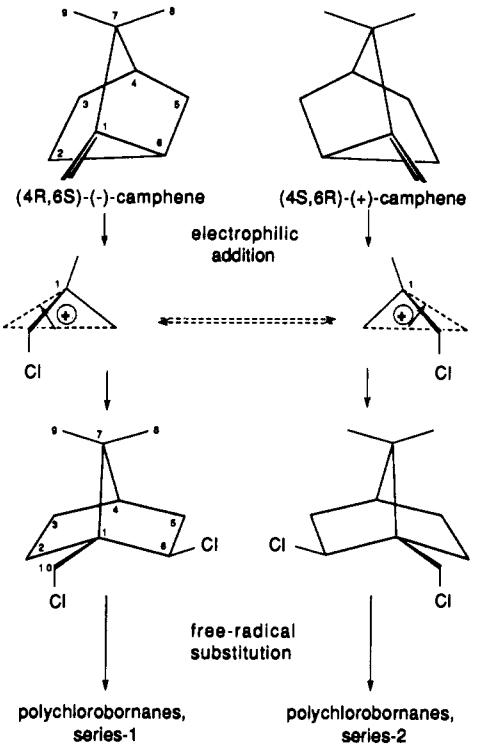
INTRODUCTION

Toxaphene (polychlorocamphene; Toxakil, Melipax, and other trade names) was a widely used, inexpensive pesticide produced by the chlorination of camphene (Sittig, 1980; Parlar, 1985; Saleh, 1991). Although its use has been drastically limited in many countries [in the United States since 1983, see Rapaport and Eisenreich (1986)], it is likely still in use in certain parts of the world (Bidleman et al., 1988; Saleh, 1991) and was, for instance, registered in former East Germany (GDR) as late as 1987–1988 (Beitz et al., 1991). Toxaphene was produced by several manufacturers in North America as well as in Europe (including Eastern Europe) and probably in other locations (Saleh, 1991). Cumulative world production is estimated to have reached 10^6 tons and still continues (Rapaport and Eisenreich, 1988). Some toxaphene components show high persistence and bioconcentration, especially in aquatic species; not surprisingly, toxaphene is now a major global environmental contaminant (Musial and Uthe, 1983; Muir et al., 1990; Parlar, 1991; Stern et al., 1992; Vetter et al., 1992).

Technical toxaphene is a complex mixture with several hundred components (theoretically tens of thousands of compounds) with the exact structure of only a few known (Saleh, 1991). Toxaphene constitutes probably the most diverse pesticide mixture, and its exact analysis is a formidable task. It appears that most components in technical toxaphene are polychlorobornanes ($C_{10}H_{18-n}Cl_n$, n = 5–12), since several of these were isolated and identified, and 2-*exo*-10-dichlorobornane is a major intermediate in the chlorination of camphene (Jennings and Herschbach, 1965). Many polychlorobornanes are chiral, but this aspect so far has received little attention.

Camphene (chiral) is a natural product occurring in different essential oils in the dextrorotatory (+)-form as well as in the levorotatory (−)-form (Windholz et al., 1976). The absolute configurations of the two enantiomers are known (Midgely et al., 1978) and shown in Chart 1: (−)-camphene has (4*R*,6*S*)-configuration, and (+)-camphene has (4*S*,6*R*)-configuration [note the different non-IUPAC numbering schemes used in Chart 1 and by Midgely et al. (1978)]. The technical production of camphene starts from

Chart 1. Proposed Mechanism for the Formation of Polychlorobornanes from Chlorination of the Two Enantiomers of Camphene via a Wagner–Meerwein Rearrangement and 2-*exo*-10-Dichlorobornane, Formulated as Electrophilic Addition and Free-Radical Substitution[a]



[a] Hydrogen atoms on carbon skeleton and on methyl substituents are not indicated. IUPAC numbering of carbon skeleton is used for the bornane system. Chiral polychlorobornanes may be formed with some retention of configuration.

α-pinene (Bartholomé et al., 1982; Saleh, 1991), but other synthetic pathways are also known. α-Pinene, the principal constituent of turpentine, is also chiral and reported to occur in the (+)-form in Greek, in the (−)-form in Spanish, and as a more or less racemic mixture in North American turpentine (Bartholomé et al., 1981). It is not

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Table 1. Technical Toxaphenes Analyzed

smpl ident	source	manufacturer (batch, date)	label
US-1	FHO[b]	Hercules, Inc., Wilmington, DE, U.S.A. (X-16189-9, 7/27/71)	Toxaphene (68.9% Cl)
GDR-2	FHO[b]	VEB Fahlberg-List, Magdeburg, GDR	Melipax (66.8% Cl)
UNK-3	RIKILT[a]	unknown	Toxaphene
UNK-4	Ehrenstorfer[c]	unknown	Camphechlor (62.7% Cl)
UNK-5	Ehrenstorfer[c]	unknown	Camphechlor (60% Cl)

[a] Obtained from W. A. Traag, Institute for Quality Control of Agricultural Products, Wageningen, The Netherlands. [b] Obtained from L. Alder/H. Beck, Federal Health Office, Berlin, Germany. [c] Obtained from Ehrenstorfer GmbH, Augsburg, Germany.

Table 2. Toxaphene Components Investigated in Technical Toxaphenes

abbrev[a]	compd, structure	achiral HRGC, ECNI	chiral HRGC, EI SRM transition[b]
TC1	heptachlorobornane	343 $(M - Cl + 2)^-$	$342^+ \rightarrow 246^+$
TC2	heptachlorobornane	343 $(M - Cl + 2)^-$	$342^+ \rightarrow 212^+$
TC3	heptachlorobornene	343 $(M - Cl + 4)^-$	$376^+ \rightarrow 246^+$
TC4	heptachloroisocamphane	343 $(M - Cl + 2)^-$	na
TC5	octachlorobornane	379 $(M - Cl + 4)^-$	$376^+ \rightarrow 280^+$ [c]
TC6	octachlorobornane	379 $(M - Cl + 4)^-$	$376^+ \rightarrow 280^+$
TC7	octachlorobornane	379 $(M - Cl + 4)^-$	na
TC8	octachlorobornane (toxicant A)	379 $(M - Cl + 4)^-$	na
TC9	octachlorobornene	379 $(M - Cl + 6)^-$	$410^+ \rightarrow 280^+$
TC10	octachloroisocamphane	379 $(M - Cl + 4)^-$	na
TOX8	octachlorobornane	379 $(M - Cl + 4)^-$	na
TOX9	nonachlorobornane	413 $(M - Cl + 4)^-$	$410^+ \rightarrow 314^+$

[a] Elution of all components indicated in Figure 5. [b] na, not analyzed by chiral HRGC. [c] Not enantiomerically resolved, chirality unknown.

known whether camphene used in the production of toxaphene has the same enantiomeric composition and whether any enantiomeric excess in camphene would be retained in the polychlorobornanes and other components formed. It is not even known how constant the exact isomeric composition of toxaphene is, particularly of products from different producers, since comparative analyses have hardly ever been reported.

In a previous study (Buser and Müller, 1994) we reported on the application of chiral high-resolution gas chromatography (HRGC) toward the enantioselective determination of some toxaphene components in a small number of aquatic environmental samples and in a technical product. Electron-capture, negative ionization (ECNI) mass spectrometry was used for congener group analysis of the various chloro compounds and electron ionization (EI) MS/MS monitoring specific ion transitions for the isomer selective detection of some key toxaphene components. Not only did the enantiomeric ratios of several components differ in the aquatic samples (likely from biotic processes encountered), but small deviations from an exact 1:1 ratio of some components were apparent in the technical mixture and could suggest technical toxaphene not to be entirely racemic. One might hypothesize that toxaphene from different producers may differ not only in isomeric but also in enantiomeric composition. It would be an exceptional finding if toxaphene from North American producers would be more or less racemic and toxaphene from European producers optically active due to the use of different α-pinene or camphene. An even more significant finding would be if environmental toxaphene, e.g., in the Arctic with inputs from North America and possibly Eastern Europe (Bidleman et al., 1987, 1989), could be traced to one or the other type of toxaphene.

In the present study we report the chemical composition of five technical toxaphenes including one of U.S. and one of GDR production. We report detailed information on the isomeric composition of toxaphene as well as on the enantiomeric composition of some key components previously characterized (Buser and Müller, 1994). Surprisingly, all five products showed remarkably similar isomeric as well as enantiomeric compositions despite the likely different production conditions used; the enantiomeric ratios for some key components in the technical toxaphene again indicated small but significant deviations from an exact 1:1 ratio for racemic mixtures. Chiroptical measurements confirmed the nonracemic character of all products with a small excess of enantiomers originating from (+)-camphene. Chlorination experiments with optically active camphenes showed some of the enantiomeric excess retained in the polychlorobornanes. Results are included from the chlorination of achiral tricyclene leading to largely the same compounds.

EXPERIMENTAL PROCEDURES

Technical Toxaphenes Analyzed. Details on the five samples of toxaphene analyzed are given in Table 1. All products had a similar appearance of waxy semisolid materials having yellowish color. The samples included a product of U.S. origin with known production date and batch number, and a sample of GDR production. They were judged to represent products of widely differing manufacturing facilities. The actual producers of the three other samples are unknown. Sample UNK-4 is the one used in a previous investigation (Buser and Müller, 1994). The products were dissolved in toluene at 100 ng/μL for ECNI MS and at 1000 ng/μL for EI MS/MS analysis. Care was taken to get representative subsamples by melting the semisolid materials (70 °C), since the enantiomeric and isomeric compositions of liquid and solid may differ.

Toxaphene Components Investigated. The key toxaphene components analyzed were previously characterized (Buser and Müller, 1993); they are listed in Table 2. Except for TOX8, TOX9, and, presumably, TC8 (toxicant A), the exact structures of these compounds are unknown. It is presently unknown whether any of these compounds correspond to the major polychlorobornanes isolated by Burhenne et al. (1993).

Monoterpene Hydrocarbons Investigated. Camphene (three samples of different enantiomeric composition) and tricyclene (structure, see Chart 2) were obtained from Aldrich (Buchs, Switzerland); supplier information is given in Table 3. According to this supplier the two (+)-camphenes (one of technical and one of purified quality) are of synthetic origin and prepared from α-pinene; (−)-camphene is reported to be of natural origin. All hydrocarbons were analyzed for isomeric and enantiomeric purity using achiral and chiral HRGC–MS (see below).

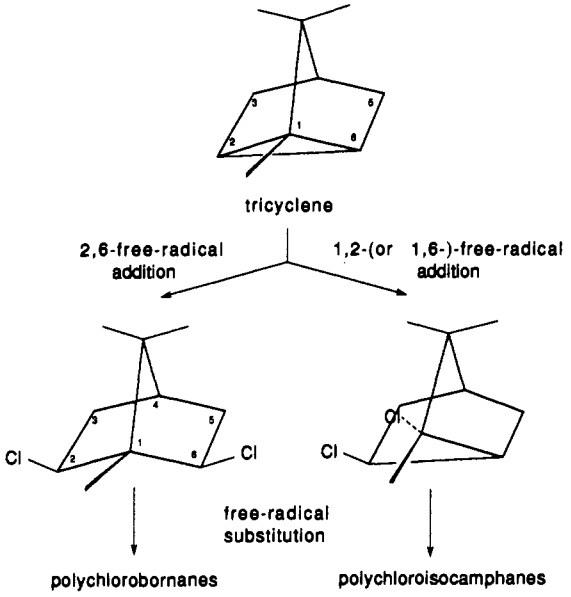
Chlorination of Camphenes and Tricyclene. The experiments were carried out with the technical (−)-camphene, the purified (+)-camphene, and tricyclene. Chlorination was carried out by passing Cl_2 from a gas cylinder through a solution of 200–300 mg of camphene or tricyclene dissolved in 2–3 mL of CCl_4. No cooling was applied, and the temperature initially rose to

Table 3. Information on the Camphenes and Tricyclene Used in the Chlorination Experiments

compd[a]	supplier information[b]	prep	comp[c] (%)	ER value[d]	EE[e] (%)
(+)-camphene	technical, 79%, remainder tricyclene, a_D = +14.8°	synthetic	81 (13)	0.74 ± 0.01	+14.9
(+)-camphene	purified, 94.4%, a_D = +20.7°	synthetic	94 (2.5)	0.67 ■ 0.01	+19.8
(−)-camphene	technical, 84.7%, a_D = −48.6°	natural	85 (<0.5)	3.85 ± 0.1	−58.8
tricyclene	99%	synthetic	(99)		

[a] All samples from Aldrich. [b] Optical rotation in ethanol, c = 4. [c] Relative amounts (%) of camphene and (in parentheses) tricyclene, as determined from total ion chromatogram data using the OV1701-BSCD HRGC column, summing the values of the two enantiomers of camphene. [d] ER values as determined by chiral HRGC using EI SIM (m/z 136) data. [e] Enantiomeric excess as calculated by EE (%) = 100 × (ER − 1)/(ER + 1), sign indicating levo- and dextrorotation.

Chart 2. Proposed Mechanism for the Formation of Polychlorobornanes and Polychloroisocamphanes from Chlorination of Achiral Tricyclene, Formulated as Free-Radical Addition and Substitution (Ionic Mechanisms Also Conceivable)[a]



[a] Chiral products will be formed as racemic mixtures independent of the reaction mechanism. Cl substituents are not necessarily *exo*-configuration.

60–70 °C. Addition of Cl_2 was continued for 8 h while the temperature remained at room temperature. Initial experiments were carried out with reaction times of 30 min. Occasionally, CCl_4 was added to compensate for volatilization losses of solvent. After reaction, the solutions were evaporated in vacuo, and the materials were dissolved in 2 mL of dichloromethane (DCM) and passed through 1 g of basic alumina (Woelm, Eschwege, Germany) in a disposable Pasteur pipet, eluting with a total of 10 mL of DCM. After concentration and reconstitution in toluene, the samples were directly analyzed, injecting aliquots corresponding to 100 (ECNI MS) or 1000 ng (EI MS/MS).

HRGC–MS Analyses. A VG Tribrid double-focusing magnetic sector hybrid mass spectrometer (VG Analytical Ltd., Manchester, England) was used under conditions similar to those reported by Buser and Müller (1994). Briefly, a 25-m SE54 fused silica column was used for achiral HRGC and a 20-m OV1701 fused silica column with 30% *tert*-butyldimethylsilyl-β-cyclodextrin (BSCD; amount relative to OV1701) for chiral HRGC. The temperature programs were as reported previously (Buser and Müller, 1994). Congener group analysis was carried out by ECNI MS using computer-reconstructed mass chromatograms from full-scan ECNI mass spectra (m/z 35–535, 1.16 s/scan; Ar/10% CH_4 as buffer gas), in particular m/z 343, 379, and 413 for the hepta-, octa-, and nonachloro compounds. Some key toxaphene components (see Table 2) were selectively detected by EI MS/MS (70 eV, 180 °C; collision gas, Ar; collision energy, 18 eV; quadrupole mass filter for daughter ion resolution) by selected reaction monitoring (SRM) using ion transitions characteristic for the compounds under investigation, in particular ion transitions involving the elimination of $C_2H_2{}^{35}Cl_2$ (96 Da) and $C_2H^{35}Cl_3$ (130 Da) from retro-Diels–Alder (RDA) fragmentations of $(M - HCl)^{\cdot+}$ and $M^{\cdot+}$ ions of polychlorobornanes and polychlorobornenes, respectively (Buser and Müller, 1994; Buser et al., 1993). Generally, the more intense $^{35}Cl_x{}^{37}Cl$ isotopomers were used.

Camphene was analyzed on a 20-m OV1701-PMCD (PMCD = permethyl-β-CD, amount 10% relative to amount of OV1701) fused silica HRGC column programmed as follows: 50 °C, 2 min isothermal, 10 °C/min to 60 °C, then 2.5 °C/min to 150 °C. Samples (1–2 ng) were injected in *n*-hexane. Full-scan EI mass spectra were recorded and selected ion monitoring (SIM; m/z 136, 121, 107, and 93) was used for analyte detection.

The denotation of enantiomers is as previously used in studies on chlordane and toxaphene components; enantiomeric ratios (ER values) and resolution (R) are defined accordingly (Buser et al., 1992; Buser and Müller, 1994). Average and range from two or more replicate measurements are reported.

Chiroptical Measurements. A polarimetric detector (Buechi Ltd., Flawil, Switzerland) was operated in a static mode using a 350-μL cell and in combination with high-performance liquid chromatography (HPLC) in a flow-through mode using a 40-μL cell. The HPLC system consisted of a Spectra-Physics (San Jose, CA) Model 8800 pump delivering a flow of 0.6 mL/min of heptane/0.1% 2-propanol. The samples were injected via a Rheodyne (Cotati, CA) Type 7125 injector with a 20-μL loop. A silica column (achiral, 250 × 3 mm, 5-μm particle size; Macherey-Nagel, Düren, Germany) eluted camphene and tricyclene after 3.1 min and the toxaphene components after 3.1–6.0 min. An Erma (Tokyo, Japan) Model 7125A refractive index (RI) detector was used to establish proper chromatographic conditions in the initial phase of the experiments.

Using the polarimetric detector, dextrorotation resulted in positive signals and levorotation in negative signals, as recorded on a strip chart recorder (1 mV equivalent to an optical rotation of 1 mdeg; reproducibilty ≈1%). Specific rotations were determined from static measurements and calculated as $[\alpha] = \alpha_m/(c \times l)$ where α_m is the optical rotation measured, c is the concentration in g/mL, and l is the length of the cell in decimeters. Samples were dissolved in *n*-heptane at concentrations of 0.5–1 mg/mL for camphene and the chlorination products and at 30 mg/mL for the commercial toxaphenes. Spiking experiments were done by adding small amounts of chlorination products from (+)- and (−)-camphene to confirm direction and extent of rotation of samples with small $[\alpha]$ values (commercial toxaphenes).

RESULTS AND DISCUSSION

Mechanistic Considerations. Toxaphene is produced by chlorination of camphene ($C_{10}H_{16}$), and the majority of products are polychlorobornanes ($C_{10}H_{18-n}Cl_n$) (Parlar, 1985, 1991; Saleh, 1991) and, to a lesser degree, other components such as polychlorobornenes ($C_{10}H_{16-n}Cl_n$) (Saleh, 1991) and polychloroisocamphanes ($C_{10}H_{18-n}Cl_n$) (Seiber et al., 1975; Buser and Müller, 1994). The reaction appears to proceed via the chiral 2-*exo*-10-dichlorobornane (electrophilic addition of Cl_2) and involves a Wagner–Meerwein rearrangement (Jennings and Herschbach, 1965; Saleh, 1991; Burhenne et al., 1993). Further chlorination, likely by a free-radical mechanism, leads to polychlorobornanes. If the reaction proceeds as outlined in Chart 1, it can be rationalized that asymmetrical products are formed with some retention of configuration. The reaction could thus lead to polychlorobornanes with one series of enantiomers from (+)-camphene and the other series from

(−)-camphene. The polychlorobornanes formed via this mechanism would contain at least one Cl each at C-2 and C-10, respectively; most polychlorobornanes identified so far belong into this group of isomers. Addition of Cl_2 to the C–C double bond in camphene without Wagner–Meerwein rearrangement leads to polychloroisocamphanes (all chiral) containing at least one Cl each at C-1 and C-10, respectively; the polychloroisocamphane isolated from toxaphene belongs to this group of isomers (Seiber et al., 1975). Again, different enantiomers could be formed from (+)- and (−)-camphene. Complete retention of configuration with both types of reaction would lead to chiral polychlorobornanes and other products with the same or the reciprocal ER value of camphene (reciprocal in the case of reversed enantiomer elution). Partial racemization as indicated in Chart 1 would yield ER values closer to unity. Other chlorination pathways involving symmetrical intermediates could lead to partially or fully racemizated products. It should be pointed out that in an acid- or radical-catalyzed rearrangement of α-pinene to technical camphene (Saleh, 1991), hydride shifts may lead to partial racemization but its extent is not known and may depend on the actual reaction conditions used.

Tricyclene ($C_{10}H_{16}$), an achiral isomer of camphene, is a natural product (Karrer, 1985) as well as a byproduct in the synthesis of camphene from α-pinene. As camphene, tricyclene reacts with Cl_2 to yield a similar complex mixture of chlorinated compounds (see later). The majority of products observed were of composition $C_{10}H_{18-n}Cl_n$ and most likely polychlorobornanes and/or -isocamphanes. The reaction likely involves addition of Cl_2 followed by free-radical substitution. Addition of Cl_2 most likely occurs at the strained cyclopropane ring formed by carbon atoms C-1, C-2, and C-6, with cleavage of a C–C bond [free-radical addition of Cl_2 to cyclopropane is known, see March (1985); ionic mechanisms are also conceivable]. Assuming free-radical addition/substitution, as outlined in Chart 2, this could proceed via two pathways leading to polychlorobornanes (cleavage of the C-2–C-6 bond) or polychloroisocamphanes (cleavage of the C-1–C-2 or C-1–C-6 bond). In these cases, the polychlorobornanes would contain at least one Cl each on C-2 and C-6, and the polychloroisocamphanes at least one Cl each at C-1, and C-2 or C-6, respectively. In both cases and independent of the mechanism, racemic mixtures of chiral products are expected. The situation thus is different from the situation with chiral camphene in which polychlorobornanes and polychloroisocamphanes may be formed as nonracemic mixtures.

Enantiomeric Composition of Camphenes. The $[\alpha]_D$ values listed for the camphenes by the supplier (−48.9, +14.8, and +20.7°) and literature values as high as −119.1° for (−)-camphene (Midgely et al., 1978) suggest a different enantiomeric purity of our three samples. This was confirmed by chiral HRGC using OV1701-PMCD. Using this column, the two enantiomers of camphene were resolved ($R \approx 2.3$) and separated from the earlier-eluting tricyclene, as shown in Figure 1. The compositions of the camphenes determined from total ion chromatograms are reported in Table 3. The technical and the purified (+)-camphene contained ≈13 and ≈2.5% tricyclene, respectively, but tricyclene was not detectable (<0.5%) in the natural (−)-camphene. Camphene and tricyclene both have the composition $C_{10}H_{16}$ but were easily distinguished by full-scan EI MS. As expected, the EI mass spectra of the two enantiomers of camphene were identical.

Enantiomeric ratios (ER values) were determined from EI SIM (*m/z* 136) data. The chromatogram of technical

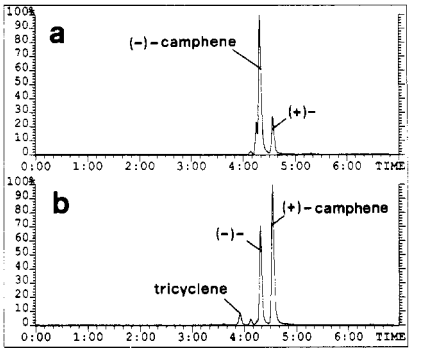


Figure 1. EI SIM (*m/z* 136) chromatograms of (a) technical (−)-camphene and (b) purified (+)-camphene, using the chiral OV1701-PMCD HRGC column. Note the separation of camphene into enantiomers with the (−)-enantiomer earlier-eluting and the (+)-enantiomer later-eluting. Note the presence of small amounts of other monoterpene hydrocarbons including tricyclene.

(−)-camphene shown in Figure 1a indicates the (−)-enantiomer as earlier-eluting (ER = 3.85 ± 0.1); the chromatogram of purified (+)-camphene shown in Figure 1b confirms the (+)-enantiomer as later-eluting (ER = 0.67 ± 0.01). Technical (+)-camphene showed an enantiomeric ratio of 0.74 ± 0.01 with a relatively higher amount of tricyclene (chromatogram not shown). The ER values reported for all three products correspond to enantiomeric excesses (EE values) of +14.9, +19.8, and −58.8%, respectively (see Table 3; sign indicates excess of dextro- and levorotatory enantiomer). A plot of the optical rotation (absolute $[\alpha]_D$ values as listed by the supplier and corrected for the content of camphene) vs the absolute EE values (as determined by chiral HRGC) showed a linear relationship, and thus the values support each other. Forcing the regression line through the origin (zero rotation for a racemic product) gave a correlation coefficient, r^2, of 0.996 and a slope of 1.005. The $[\alpha]_{max}$ value calculated for 100% enantiomeric excess is 100.5 ± 3.8° (mean, standard error). The data indicate neither product to be racemic or enantiomerically pure.

Chlorination of Camphene and Tricyclene. Chlorination of (−)- and (+)-camphene led to complex mixtures with dozens of compounds. As indicated by the *m/z* values of $(M - Cl)^-$ ions in the ECNI mass spectra, the majority of components have the composition $C_{10}H_{18-n}Cl_n$ and thus correspond to polychlorobornanes or -isocamphanes. They were predominantly hexa- and heptachlorinated. In Figure 2 we show ECNI mass chromatograms *m/z* 343, 379, and 413 for the hepta-, octa-, and nonachloro compounds originating from (−)- and (+)-camphene and analyzed on the achiral SE54 HRGC column. The chromatograms of both reaction products indicate a very similar composition with almost identical isomer profiles and only minor differences in the relative abundances of some isomers (note the presence of components TC1, TC2, TC5, TC6, TC7, TC8, and TOX9 as indicated). Apparently, the different impurities in (−)- and (+)-camphene have little effect on the composition of these reaction products. Both chromatograms are of similar complexity as those of the technical products (see later), with largely the same congeners present, particularly the polychlorobornanes, but detailed analyses of the laboratory products revealed the absence of TC3 and TC9 (polychlorobornenes) and a lower abundance of some other components such as TC5. Less stringent reaction conditions in the laboratory than in the technical processes likely account for the smaller abundance of higher chlorinated compounds such as TOX9 and other nonachlorobornanes.

ECNI mass chromatograms of the tricyclene reaction

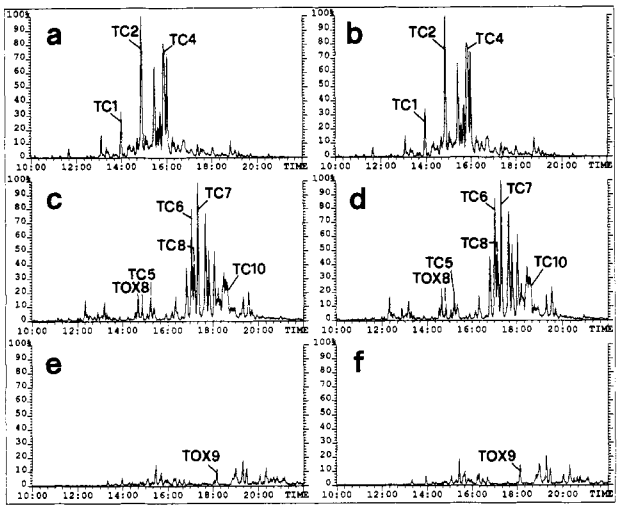


Figure 2. ECNI mass chromatograms showing elution of (a, b) hepta- (m/z 343), (c, d) octa- (m/z 379), and (e, f) nonachloro compounds (m/z 413) from the chlorination of technical (−)-camphene (left-side panels) and purified (+)-camphene (right-side panels), using the achiral SE54 HRGC column. Note the almost identical isomer profiles in both products. See text for abbreviations. Relative intensities are normalized to that of the octachloro compounds (m/z 379), with vertical expansions of 0.25× for heptachloro and 2× for nonachloro compounds.

product showed a similar complex pattern but with a higher proportion of earlier-eluting components (data not shown). The majority of products have a composition of $C_{10}H_{18-n}Cl_n$ (polychlorobornanes or -isocamphanes) and thus are formed by addition of Cl_2 and subsequent substitution (see above). The chromatograms suggest the presence of TC1, TC2, TC5, and TC6, whereas TOX9 was absent. Only a few components have a composition of $C_{10}H_{16-n}Cl_n$ (polychlorotricyclenes, polychlorobornenes); TC3 and TC9 (polychlorobornenes) were not among these products. Since tricyclene is a byproduct in technical camphene and its chlorination leads to an equally complex mixture, it may account for some of the components in technical toxaphene but probably not for the major ones.

The chlorination products were analyzed using the chiral OV1701-BSCD HRGC column and EI SRM. This column separates some toxaphene components enantiomerically, but others remained unresolved [see Table 2 and Buser and Müller (1994)]. In Figure 3 we show SRM chromatograms selective for TC1, TC6, and TOX9 (hepta-, octa-, and nonachlorobornanes, respectively). TC5, another octachlorobornane, is also detected but not enantiomerically resolved; it is unknown whether this component is actually chiral.

The EI SRM chromatograms in Figure 3a–c of the chlorination product of (−)-camphene show the presence of both enantiomers of these components but with a significant excess of enantiomer TC1-1 (ER = 1.87), TC6-1 (ER = 1.82), and TOX9-2 (ER = 0.45; reciprocal value 2.22). The chlorination product of (+)-camphene (see Figure 3d–f) showed a reversed behavior with a significant excess of enantiomers TC1-2 (ER = 0.56) and TC6-2 (ER = 0.73). In the case of TOX9, the abundance was too low to give an accurate ER value (estimated ER ≈ 1.1; reciprocal value ≈ 0.9). The chromatograms clearly show that one enantiomer of these components is formed preferably from (−)-camphene and the other from (+)-camphene. The results thus show that some of the enantiomeric excess is retained in the products and that the chlorinations thus proceed with some retention of configuration. Furthermore, the data show that the elution orders of enantiomers of products and parent compounds

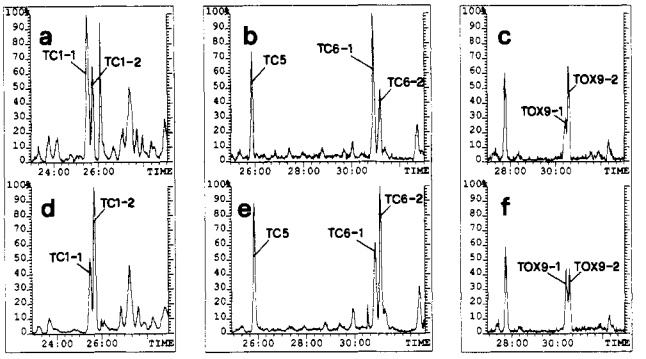


Figure 3. EI SRM chromatograms of the chlorination products of technical (−)-camphene (a–c) and purified (+)-camphene (d–f), using the chiral OV1701-BSCD HRGC column. (a, d) SRM chromatogram $342^+ \rightarrow 246^+$ showing elution of TC1 (a heptachlorobornane); (b, e) SRM chromatogram $376^+ \rightarrow 280^+$ showing elution of TC5 and TC6 (octachlorobornanes); (c, f) SRM chromatograms $410^+ \rightarrow 314^+$ showing elution of TOX9 (a nonachlorobornane). Note the resolution of TC1, TC6, and TOX9 into enantiomers and their formation as nonracemic mixtures from (−)- and (+)-camphene.

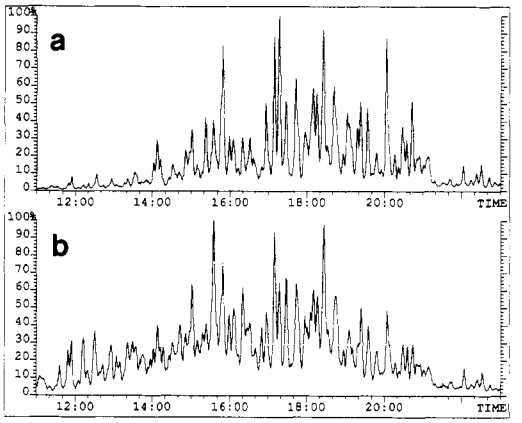


Figure 4. EI SIM (m/z 159) chromatograms showing elution of penta- to decachloro compounds (polychlorobornanes and others) in (a) technical toxaphene US-1 and (b) technical toxaphene GDR-2, analyzed on the achiral SE54 HRGC column. Note the wider distribution of chloro compounds in GDR-2, but with largely the same components present in both samples.

are not necessarily identical. The sterical influence in enantiomers of different isomers apparently is different.

The enantiomeric excesses as indicated by the ER values of these components generally were lower than those of (−)- and (+)-camphene, suggesting some racemization during chlorination. In the case of TC6 from (−)-camphene the ER values (1.82 and 3.85, respectively) indicate ≈50% racemization.

Analysis of Technical Toxaphenes. Isomer and congener group analysis of the five technical materials was carried out by EI and ECNI MS using the achiral SE54 HRGC column. In Figure 4 we show EI SIM chromatograms (m/z 159; dichlorotropylium ion, $C_7H_5Cl_2^+$) of two of the products analyzed. The chromatograms are relatively nonselective for toxaphene components. Except sample GDR-2, all products showed a very similar composition; sample GDR-2 had a somewhat wider distribution with some additional earlier-eluting components (see Figure 4b). In Figure 5 we show ECNI mass chromatograms (m/z 343, 379, and 413) for the hepta-, octa-, and nonachloro compounds in samples US-1 and GDR-2. Corresponding chromatograms of the two products are very similar, with only minor differences in the relative abundance of some components. For instance, sample GDR-2 showed a lower abundance of TC5, TC8,

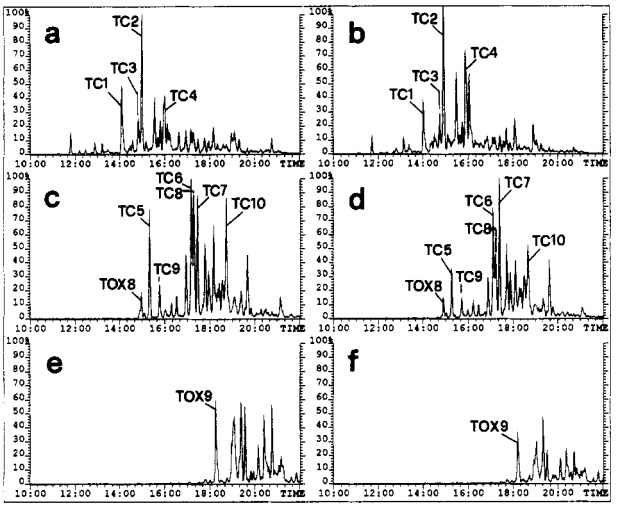


Figure 5. ECNI mass chromatograms showing elution of (a, b) hepta- (m/z 343), (c, d) octa- (m/z 379), and (e, f) nonachloro compounds (m/z 413) in technical toxaphene US-1 (left-side panels) and technical toxaphene GDR-2 (right-side panels), using the achiral SE54 HRGC column. Note the very similar isomer profiles of the U.S. and GDR products. See text for abbreviations. Relative intensities are normalized to that of the octachloro compounds (m/z 379).

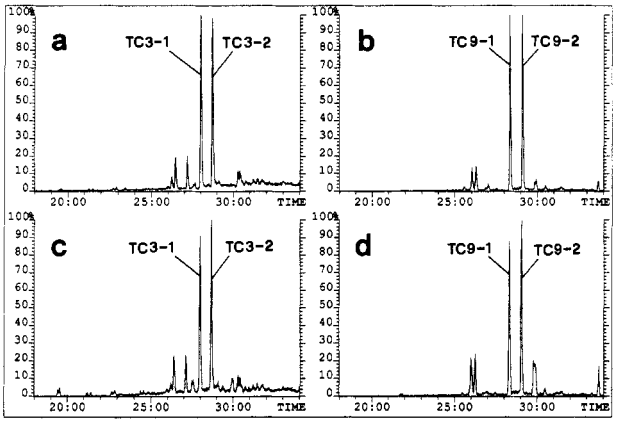


Figure 6. EI SRM chromatograms $376^+ \rightarrow 246^+$ (a, c) and $410^+ \rightarrow 280^+$ (b, d) showing elution of heptachlorobornene TC3 and octachlorobornene TC9, respectively, in technical toxaphenes, using the chiral OV1701-BSCD HRGC column. (a, b) Technical toxaphene US-1; (c, d) technical toxaphene GDR-2. Note the separation of TC3 and TC9 into enantiomers and the very similar isomer/enantiomer profile of the U.S. and GDR products.

and nona- and higher chlorinated compounds. All samples showed the presence of the 12 key components, including the polychlorobornenes (TC3 and TC9) and the polychloroisocamphanes (TC4 and TC10).

The enantiomeric composition of several key components was determined using chiral HRGC. In Figure 6 we show EI SRM chromatograms $376^+ \rightarrow 246^+$ and $410^+ \rightarrow 280^+$ selective for hepta- and octachlorobornenes TC3 and TC9, respectively. These components were detected in all five samples, as shown for samples US-1 and GDR-2, and resolved into pairs of enantiomers ($R \approx 4.0$) with the two pairs closely eluting but clearly separated. The chromatograms indicate racemic mixtures for both components in all of the products (see Table 4). Additional minor peaks are visible, particularly in sample GDR-2; several doublets suggest the presence of additional chiral compounds.

In Figure 7 we show EI SRM chromatograms $342^+ \rightarrow 246^+$ and $342^+ \rightarrow 212^+$ selective for the heptachlorobornanes TC1 and TC2, respectively. All five samples showed very similar chromatograms and similar distributions of

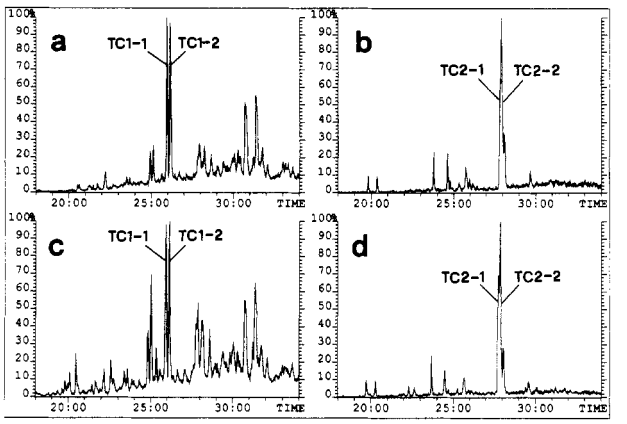


Figure 7. EI SRM chromatograms $342^+ \rightarrow 246^+$ (a, c) and $342^+ \rightarrow 212^+$ (b, d) showing elution of heptachlorobornanes TC1 and TC2, respectively, in technical toxaphenes, using the chiral OV1701-BSCD HRGC column. (a, b) Technical toxaphene US-1; (c, d) technical toxaphene GDR-2. Note the very similar isomer/enantiomer profile of the U.S. and GDR products. Note the separation of TC1 and the partial separation of TC2 into enantiomers; enantiomer TC2-2 coelutes with another component.

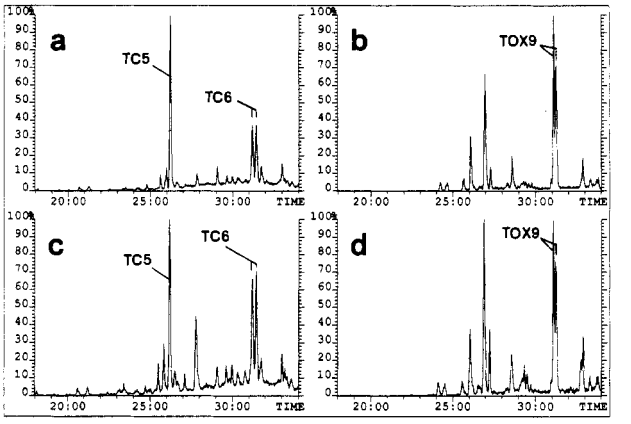


Figure 8. EI SRM chromatograms $376^+ \rightarrow 280^+$ (a, c) and $410^+ \rightarrow 314^+$ (b, d) showing elution of octachlorobornanes TC5 and TC6 and nonachlorobornane TOX9, respectively, in technical toxaphenes, using the chiral OV1701-BSCD HRGC column. (a, b) Technical toxaphene US-1; (c, d) technical toxaphene GDR-2. Note the very similar isomer/enantiomer profile of the U.S. and GDR products. Note the separation of components TC6 and TOX9 into enantiomers.

Table 4. Enantiomeric Ratios (ER Values) of Chiral Toxaphene Components in Five Technical Toxaphenes

component	sample				
	US-1	GDR-2	UNK-3	UNK-4	UNK-5
TC1	0.94 ± 0.02	0.97 ± 0.03	0.92 ± 0.02	0.92 ± 0.02	0.94 ± 0.02
TC3	0.99 ± 0.01	0.99 ± 0.01	0.96 ± 0.03	0.99 ± 0.03	1.02 ± 0.02
TC6	0.98 ± 0.02	1.00 ± 0.05	1.05 ± 0.02	1.04 ± 0.03	1.00 ± 0.02
TC9	0.98 ± 0.01	0.96 ± 0.03	1.02 ± 0.02	1.00 ± 0.03	1.03 ± 0.03
TOX9	1.16 ± 0.02	1.08 ± 0.02	1.11 ± 0.03	1.13 ± 0.02	1.00 ± 0.01

enantiomers and isomers. As shown for samples US-1 and GDR-2, component TC1 is clearly separated into a pair of enantiomers (see Figure 7a,c), whereas TC2 apparently coelutes with another component (see Figure 7b,d). In the case of TC1, the ER values suggest a somewhat lower abundance of the earlier-eluting enantiomer (see Table 4). The chromatograms indicate the presence of additional components in both products but with a higher abundance in GDR-2.

In Figure 8 we show EI SRM chromatograms $376^+ \rightarrow 280^+$ and $410^+ \rightarrow 314^+$ selective for some octa- and nonachlorobornanes, respectively, and illustrated for samples US-1 and GDR-2. In Figure 8a,c the elution of

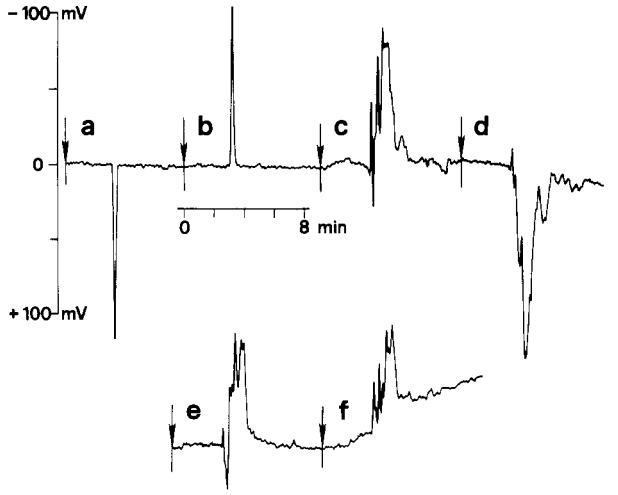


Figure 9. Chiroptical (achiral) HPLC chromatograms of (a, b) (+)- and (−)-camphene, (c, d) the chlorination products of (+)- and (−)-camphene, and (e, f) technical toxaphenes US-1 and GDR-2, respectively. Note the changed direction of rotation of (+)- and (−)-camphene upon chlorination, and the small net levorotation of the technical toxaphenes. Negative signals (upward) correspond to levorotation, positive signals (downward) to dextrorotation.

Table 5. Chiroptical Data of Camphene, Tricyclene, and Chlorination Products and Commercial Toxaphenes

compd/sample	[α] static (deg)	sample	[α] static (deg)
(−)-camphene	−89.6	US-1	−0.95
(−)-camphene, chlorinated, 30 min	+32.7	GDR-2	−0.50
(−)-camphene, chlorinated, 8 h	+27.4	UNK-3	−1.30
(+)-camphene	+40.1	UNK-4	−0.45
(+)-camphene, chlorinated, 8 h	−35.4	UNK-5	−0.30
tricyclene	<0.05		
tricyclene, chlorinated, 8 h	<0.05		

TC5 and TC6 is shown; TC6 is resolved enantiomerically, and the ER values suggest its presence as nearly racemic in all five samples (see Table 4). TC5 is eluted as a single peak; TOX8, another octachlorobornane and a minor component in these technical products, was not enantiomerically resolved. In Figure 8b,d the elution of TOX9 and its resolution into enantiomers are shown. The ER values of all five samples ranged from 1.00 to 1.16 (see Table 4), indicating small deviations from an exact 1:1 ratio in some of the samples.

Chiroptical Data. Chiroptical measurements with (+)-camphene (purified) and (−)-camphene (technical) showed levo- and dextrorotation as indicated by large negative and positive peaks in HPLC chromatograms, respectively (see Figure 9a,b). The [α] values for camphenes as determined by the static method were −89.6 and +40.1°, respectively (see Table 5). The absolute values are larger than those listed by the supplier, possibly due to the different experimental conditions (solvent, concentration, wavelength), but still proportional. Tricyclene showed no optical rotation ([α] <0.05°).

Chiroptical measurements of the chlorination products of (+)- and (−)-camphene showed a change in the direction of rotation as compared to the parent compounds. The net rotation of the 8-h chlorination product of (+)-camphene was levorotatory with an [α] value of −35.4°. The net rotation of the (−)-camphene products was dextrorotatory with [α] values of +32.7 and +27.4° for the 30-min and 8-h chlorination products, respectively. Chiroptical HPLC chromatograms confirmed the changed direction of rotation by showing a broad negative peak envelope for the chlorination product of (+)-camphene and a broad positive peak envelope for the chlorination product of (−)-camphene (see Figure 9c,d). The chlorination product of tricyclene showed no net rotation ([α] <0.05°).

Chiroptical measurements showed a small net rotation which was levorotatory for all five commercial toxaphenes ([α] values −0.3 to −1.3°, see Table 5). This indicates a small enantiomeric excess of (+)-camphene in the synthesis of toxaphene and confirms the nonracemic character of all these products. Chiroptical HPLC chromatograms confirmed the levorotation of all five products by the negative peak envelopes, as shown for samples US-1 and GDR-2 in Figure 9e,f.

CONCLUSIONS

The five commercial toxaphenes studied had remarkably similar isomeric as well as enantiomeric compositions despite the fact that at least two of the samples originate from widely different production facilities. Despite the complex reaction pathways, chlorination seems to be quite reproducible and leads to largely the same components. The small differences in composition among the different products are likely not sufficiently large to actually trace environmental toxaphene to one or the other product.

Most of the components appear to be polychlorobornanes or -isocamphanes, as indicated by the composition $C_{10}H_{18-n}Cl_n$ deduced from the ECNI mass spectra. Some components with a composition $C_{10}H_{16-n}Cl_n$ were assigned to polychlorobornenes (components TC3 and TC9). It was suggested that such compounds could be polychlorotricyclenes (Parlar, 1991). Whereas the chlorination of tricyclene in our model reaction actually led to a vast number of polychloro compounds that were not fully analyzed, TC3 and TC9 were clearly not among them.

Laboratory-scale chlorination of (−)- and (+)-camphene led to polychlorobornanes with some retention of configuration. Therefore, nonracemic mixtures of some chiral products may be expected in the synthesis of toxaphene from enantiomerically enriched camphene. Preliminary results show that some of these compounds are formed from achiral tricyclene, which may be present in synthetic camphene. In this case racemic mixtures are expected. Unfortunately, we have presently no information on the actual composition (isomeric, enantiomeric) of camphene used in the production of toxaphene.

ER values of five key components (three polychlorobornanes, two polychlorobornenes) were determined and found to be very similar in all commercial products, some showing small but definite deviations from an exact 1:1 ratio. The largest value of 1.16 thus determined (TOX9 in the U.S. product) indicates an enantiomeric excess of ≈7.5% of the first-eluting enantiomer, originating from (+)-camphene. At a ≈50% racemization level, as in the laboratory-scale chlorinations, this indicates an enantiomeric excess of ≈15% of (+)-camphene in the synthesis. This excess, according to our model reactions, would lead to ER values <1.00 for TC1 and TC6, because of a reversed elution of enantiomers of these components. While this is observed for TC1 (ER values 0.92–0.97), it is not observed for TC6 (ER values 0.98–1.03). Since components may originate via different reaction pathways, and possibly even from different precursors (camphene, tricyclene), ER values must not necessarily be identical for all components.

Chiroptical measurements confirmed the nonracemic nature of all these products. Interestingly, the direction of rotation of polarized light of the camphenes was changed upon chlorination: the net rotation of the chlorination products of (−)-camphene was dextrorotatory, while that

of the chlorination products of (+)-camphene was levorotatory. The chlorination products of tricyclene, which include polychlorobornanes and -isocamphanes, showed no net rotation. All five commercial products showed small net rotations which were levorotatory, confirming small enantiomeric excesses of (+)-camphene in their syntheses. The actual enantiomeric excesses are difficult to estimate from these data since levo- and dextrorotatory components may lead to some compensation. Similarly, the net rotations measured in the static mode must not necessarily agree with those measured by HPLC, since in this case compensation may be lower due to some component separation.

The data reported here are important to environmental studies on toxaphene since small deviations from enantiomeric ratios of 1:1 are thus not necessarily due to biotic processes but may result from the nonracemic technical products used. Nevertheless, the enantiomeric ratios for some key components in environmental samples deviated more from unity than for the technical mixtures analyzed. For instance, TOX9 in Antarctic penguin (*Pygoscelis adelis*) showed an ER value of 1.34 (Buser and Müller, 1994), suggesting some enantioselective biodegradation of this important recalcitrant component.

ACKNOWLEDGMENT

We thank L. Alder and H. Beck, Federal Health Office, Berlin, Germany, for technical toxaphene of U.S. and GDR production and detailed information. We gratefully acknowledge a further sample from W. A. Traag, State Institute for Quality Control of Agricultural Products, Wageningen, The Netherlands. Furthermore, we thank Buechi Ltd., Flawil, Switzerland, and the Cantonal Laboratory, Zurich, for the loan of the chiroptical and RI detectors.

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Received for review July 26, 1993. Revised manuscript received October 25, 1993. Accepted November 29, 1993.[⊗]

[⊗] Abstract published in *Advance ACS Abstracts*, January 15, 1994.