Facile Route to the Resolution of the Enantiomers of 1-Chloro-2-[2,2,2-trichloro-1-(4-chlorophenyl)ethyl]benzene (*o,p'*-DDT)

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The estrogenic component of technical grade DDT, (±)-*o,p'*-DDT, has been resolved into its enantiomeric forms. Aromatic mononitration was followed by reduction to the amine and conversion to the (+)-camphor-10-sulfonate salts. While fractional crystallization of the diastereomeric salts from acetonitrile led to optical purification of the (−)-amine salt, a reflux technique using acetonitrile provided a more expedient route to the isolation of not only the (−)-amine salt but the (+)-amine salt as well. Deamination of the resolved amines by diazonium salt formation yielded *o,p'*-DDT with specific rotations of −17.9 and 17.7°. This route may be useful for the resolution of medically important (±)-*o,p'*-DDD.

It has been established repeatedly that (±)-*o,p'*-DDT (1-chloro-2-[2,2,2-trichloro-1-(4-chlorophenyl)ethyl]-benzene) (I) is estrogenically active in both avian and mammalian systems (Kupfer, 1975). Recently the optical resolution of *o,p'*-DDT has been carried out (McBlain and Wolfe, 1975) and it has been reported that the enantiomeric forms of this compound differ in their estrogenic activities (McBlain et al., 1976). The lack of an active functional group on the *o,p'*-DDT molecule has not allowed for a direct resolution of the compound but in a previous communication we outlined an expedient route (Figure 1) to the isolation of its optical isomers (McBlain and Wolfe, 1975). In this paper we present the detailed procedures for this resolution.

MATERIALS AND METHODS

Purification of (±)-*o,p'*-DDT (I). The relative expense of pure (±)-I led to the isolation of the compound from technical grade DDT. The technical grade material was generously supplied by M. Sobelman of the Montrose Chemical Corporation of California and consisted of about 18.5% (±)-I by gas–liquid chromatographic (GLC) analysis, with the major component being *p,p'*-DDT (1,1'-(2,2,2-trichloroethylidene)bis(4-chlorobenzene)).

The GLC used was a Varian Aerograph Model 600-D operated at 185 °C with a 1.5 m × 3.2 mm o.d. Pyrex glass column packed with a 1:1 mixture of 10% DC200 and 15% QF-1 on Anakrom ABS (60–80 mesh). The carrier gas was N_2 and the detector a 250-μCi tritium source electron capture (EC) system. DDT standards were obtained from the U.S. Environmental Protection Agency (EPA). Fractional crystallization of 9 kg of the technical grade DDT from ethanol, pentane, and methanol (Haller et al., 1945) led to 516.9 g of (±)-*o,p'*-DDT containing about 1.3% *p,p'*-DDT. This purity compared favorably with that of supposedly 99+% pure commercial preparations of (±)-I where our GLC analyses revealed *p,p'*-DDT contamination ranging from 2.0 to 2.5% in three individual samples. No attempt was made to improve our yield of racemic I because of the ease of preparing such large quantities. Analysis on a Perkin-Elmer 141 photoelectric polarimeter revealed that the isolated *o,p'*-DDT was racemic. The melting points reported in the Experimental Section are uncorrected and the elemental analyses were carried out in the Department of Chemistry at the University of Alberta.

Mononitration of (±)-*o,p'*-DDT (I). I was mononitrated using a modification of the method of Sparks (1966) for chlorobenzene. The resultant reaction mixture contained five products as well as the parent compound (Figure 2). Monitoring the progress of the nitration reaction on TLC revealed that products II (1-chloro-2-[2,2,2-trichloro-1-(4-chlorophenyl)ethyl]-4-nitrobenzene) and ii (Figure 2) were the first to appear and were followed by compound iv and finally small amounts of compounds iii and v of Figure 2. From NMR spectra the unique resonance of the ortho proton on the ortho Cl ring of *o,p'*-DDT (McKinney et al., 1974) was an excellent indicator of the presence and position of substitutions on this ring (Figure 3). The parent compound (I) and the desired product (II) have been rigorously identified but the other proposed structures of Figure 2 are supported by NMR spectra only and are not definitive. It would seem that the aromatic ortho,para-directing chlorines may act in conjunction with the meta-directing trichloroethane moiety (electron withdrawing (Abou-Donia, 1975)) to determine the sites of nitration of I.

Stronger nitration conditions (more H_2SO_4) or additional reaction time resulted in an increased production of the dinitro compound (compound iv in Figure 2) previously described by Haller et al. (1945) and Forrest et al. (1946). Ideally the nitration reaction should have been stopped when the probability of mononitrating the ortho Cl ring of I to produce II equaled the probability of mononitrating the para Cl ring of II to produce iv. The yield of II from the nitration of four 50-g batches of I was 39.3% but 28.6% of I was recovered from the reaction. Obviously our 200 g of (±)-*o,p'*-DDT could have been nitrated for a longer period of time to have yielded more of the desired (±)-NO_2-*o,p'*-DDT (II) and less unaltered starting material as found for the 1-g test run of Figure 2.

Reduction of (±)-NO_2-*o,p'*-DDT (II). The method for the reduction of 1-chloro-2-[2,2,2-trichloro-1-(4-chlorophenyl)ethyl]-4-nitrobenzene (II) to its corresponding amine (1-chloro-2-[2,2,2-trichloro-1-(4-chlorophenyl)-ethyl]-4-aminobenzene) (III) (Figures 1 and 3) was similar to that for reducing nitrobenzene to aniline (Vogel, 1967). It was, however, necessary to add ethanol for solubilization of II for the reduction reaction and to add $CHCl_3$ to the resulting reaction mixture in order to solubilize the amine–chlorostannate salt complex for salt hydrolysis by the NaOH. The dried ethereal extract of III was an oil which could be crystallized from petroleum ether in an 87.0% yield (1-g reduction).

Salt Formation. Because of the high yield of the reduction reaction and the ease of oxidation of exposed III,

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Table I. Progress of the Resolution of (−)-NH_2-*o,p'*-DDT from 38.3 g of (±)-NH_2-*o,p'*-DDT-(+)-camphor-10-sulfonate Salt by Fractional Crystallization from CH_3CN

Crystallization no.	CH_3CN, ml	Salt yield, g	Time, h	$[\alpha]^{RT}D$,[a] deg	
				Salt	Amine (III)
1	1250	8.5	15	0.7	−7.1
2	120	3.8	24	3.1	−14.9
3	120	2.0	17		−30.0
4[b]	1000	15	45	−43.0	−22.9
5	500	6	11	−62.9	−69.3
6	340	4	24	−152.2	−119.6
7	240	1.5	40	−117.6	−168.7
8	100	0.5	66	−63.6	−168.0

[a] RT = room temperature. For these routine rotation determinations the temperatures of the solutions were not standardized. [b] Salts from the above crystallizations were recombined and seeded with (−)-amine salt with $[\alpha]^{RT}D$ −30.0° for the amine.

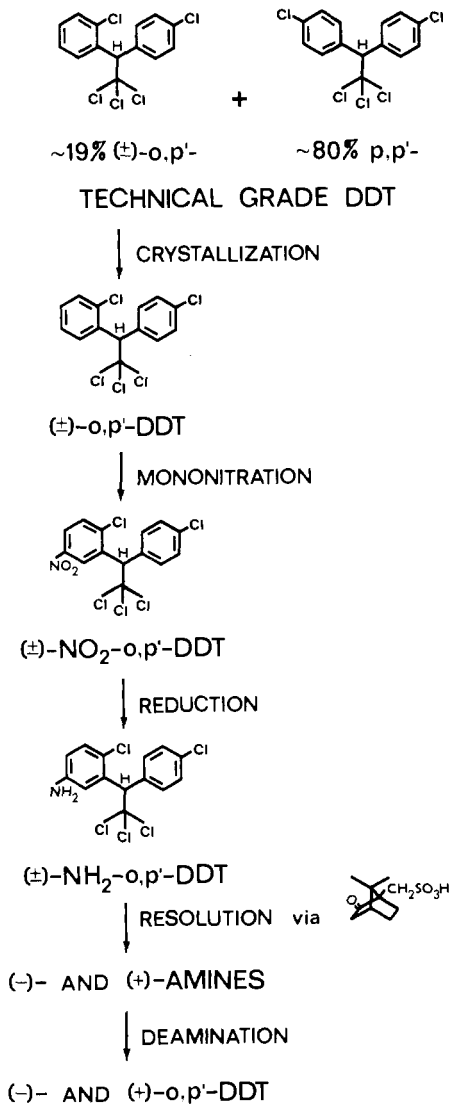

Figure 1. Route for the resolution of the enantiomers of *o,p'*-DDT (I) via a mononitrated derivative.

we combined the ethereal extract of the above reduction reaction with a CH_3CN solution containing the appropriate amount of (+)-camphor-10-sulfonic acid. The quantity of acid used was a 1:1 molar ratio to III assuming a 100% yield of III from II, presumably giving a small excess of the acid for the salt formation. Evaporation of the solvents yielded the (+)-camphor-10-sulfonate salts of (−)- and (+)-NH_2-*o,p'*-DDT (III) which were fractionally crystallized from CH_3CN or submitted to a fractional reflux technique.

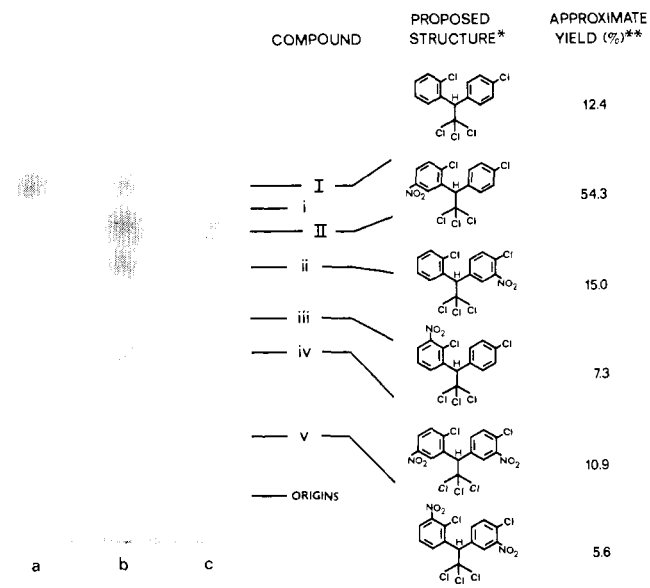

Figure 2. Thin-layer chromatogram for (a) (±)-*o,p'*-DDT (I), (b) the reaction mixture resulting from the mononitration of (±)-*o,p'*-DDT (I), and (c) (±)-NO_2-*o,p'*-DDT (II). The adsorbent was silica gel G developed with 20% diethyl ether in hexane. Compound i was an oil consisting of *o,p'*-DDT, *p,p'*-DDT, plus several other compounds (GLC) and made up about 3.3% of the reaction mixture: *, see text; **, by weight (based on a 1-g test run with compounds recovered from TLC plates).

Fractional Crystallization of the NH_2-*o,p'*-DDT-(+)-camphor-10-sulfonate salts. Our initial resolution of the enantiomers of III was carried out via the classical technique of fractional crystallization of the diastereomeric salts from an appropriate solvent (Boyle, 1971; Wilen, 1971). Typically the progress of resolutions of this type is monitored by measuring changes in (1) the specific rotation of the salt being isolated, (2) the specific rotation of the amine being isolated, or (3) the melting point of the salt being isolated. The resolution is considered complete when the above parameters cannot be altered further by continued treatment. As can be seen in Table I, the specific rotations of the salts were unreliable indicators for the progress of our resolution process so the rotations of small amounts of regenerated amines were used routinely to follow this preliminary resolution.

Before measuring the rotations of the amines it was essential that they be purified on a 4 cm × 1 cm o.d. acid-washed alumina column eluted with 20% diethyl ether in hexane in order to remove any brown (oxidized) material or unhydrolyzed salts.

Since crystallizations 1 to 3 of Table I revealed only a modest improvement of the (−)-amine rotation a small

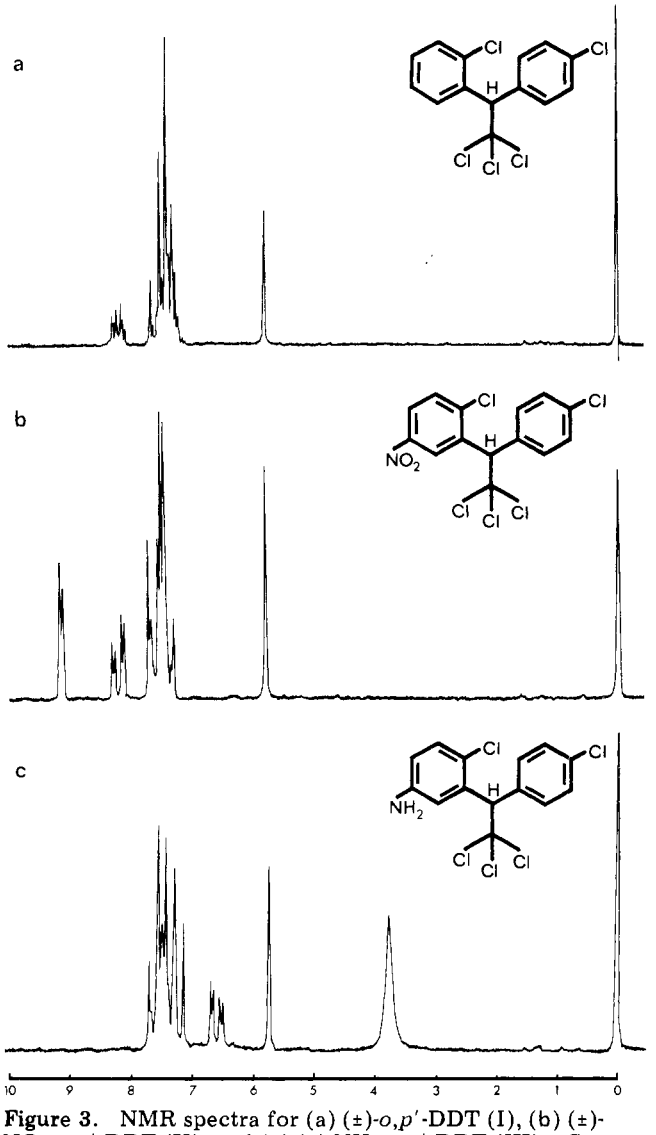

Figure 3. NMR spectra for (a) (±)-*o,p'*-DDT (I), (b) (±)-NO_2-*o,p'*-DDT (II), and (c) (±)-NH_2-*o,p'*-DDT (III). See text for description. Instrumentation: Varian Anaspect EM-360 NMR spectrometer.

amount of (−)-amine salt ($[\alpha]^{RT}_D$ −30.0° for the amine) was set aside and the remaining salts recombined. Use of the −30.0° amine salt for seed crystals led to a relatively rapid purification of the (−)-amine salt (crystallizations 7 and 8 of Table I).

Fractional Reflux of the NH_2-*o,p'*-DDT-(+)-camphor-10-sulfonate Salts. A reflux technique for the separation of diastereomeric salts has been suggested by Wilen (1971) and its use in this case was regulated by qualitative rather than quantitative means. That is, because this method of resolution was so simple and effective, the amounts of salts, solvent, and times of reflux to give various degrees of improvement in optical purity of the amines were not calculated.

To purify the levorotatory amine, (±)-amine salt or salt with a predominance of (−)-amine was dissolved in boiling CH_3CN. The volume of CH_3CN was lowered by distillation until salt was seen precipitating. At this point the distillation was stopped as a large amount of salt would precipitate while the CH_3CN continued to reflux. A small volume of CH_3CN was added to the reflux flask and the solution was kept under reflux for 2–4 h. Larger volumes of CH_3CN added to the flask (but not giving complete dissolution of the precipitated salt) and/or longer reflux times improved the resolution of the (−)-amine. Three such serial reflux treatments produced a salt with a (−)-amine (III) rotation of $[\alpha]^{25}_D$ −170.1° unimproved by further refluxing and similar in rotation to the (−)-amine previously isolated above by the classical crystallization procedure.

Our use of a double withdrawal crystallization technique yielded salts with little or no improvement of enantiomeric purity except, of course, in the case of the primary crystallization presented in Table I. That is, all other isolated salts contained amines exhibiting low dextrorotatory rotations indicating that (−)-amine salts could not be crystallized from salts containing an excess of the (+)-amine. To purify the (+)-amine, (+)-amine-enriched salts recovered from the reflux mother liquors above were dissolved in boiling CH_3CN. The volume of CH_3CN was reduced by distillation until a large amount of salt precipitated. This salt was recovered without further treatment and revealed an improved (+)-amine rotation. Six such treatments produced almost pure (+)-NH_2-*o,p'*-DDT (III).

Furthermore, we found that during this resolution the increasing melting points of the salts were excellent indices of improvements in the enantiomeric purity of the amines.

Lanthanide Shift Reagent. An attempt was made to determine the optical purity of the resolved amines utilizing a lanthanide shift reagent, Sievers' reagent or Eu(fod)$_3$ (Norell Chemical Co.). By use of Varian Anaspect EM-360 (60 MHz) and Perkin-Elmer R-32 (90 MHz) NMR spectrometers it was hoped that this reagent might split the signal for the benzylic proton of racemic NH_2-*o,p'*-DDT (III). To 50 mg of (±)-III in 0.4 ml of CCl_4 the Eu(fod)$_3$ was added in 10-mg increments and later 100-mg increments up to a total of 980 mg of the reagent. While large shifts occurred for all proton signals there was no observable split in the benzylic proton signal possibly because of the distance between the amine group and the asymmetric center of this molecule.

Deamination of (−)- and (+)-NH_2-*o,p'*-DDT (III) to (−)- and (+)-*o,p'*-DDT (I). The resolved amines were deaminated by treating their diazonium salts with hypophosphorus acid (Sager et al., 1972). The reaction solution was allowed to stand 72 h by which time it had cleared and the *o,p'*-DDT (I) enantiomers could be extracted with pentane. The specific rotations of the resolved enantiomers of I were $[\alpha]^{25}_D$ −17.9 and 17.7°. X-ray crystallographic analysis of (−)-*o,p'*-DDT has revealed that it possesses the *R* configuration (Smith and Bennett, 1977).

DISCUSSION

The route outlined above for the resolution of the enantiomeric forms of *o,p'*-DDT is both rapid and simple. While we have been unable to estimate the enantiomeric purity of the resolved compounds by NMR, the similarity of the (−) and (+) specific rotations for both the amines and parent compounds would suggest that the resolution may be complete at least for the levorotatory form. The reflux technique for this resolution has obvious advantages over the more usual crystallization method with respect to both the amount of time and number of treatments required. Our experience indicates, however, that more labile salts such as those of carboxylic acids and our aromatic amines may not withstand the rather harsh conditions of the reflux system although the choice of solvent may be critical.

The aromatic mononitration of *o,p'*-DDT yields a predominance of one derivative. If (±)-*o,p'*-DDD can be mononitrated in a similar fashion the resolution of this medically important compound (Straw and Hart, 1975) may be effected by the above route for *o,p'*-DDT.

While this resolution has obvious significance to studies of the physiological effects of *o,p'*-DDT, its significance to environmental studies is unknown because the environmental fate of the racemic compound has not been delineated. Technical grade DDT continues to be an important antimalarial agent and since (±)-*o,p'*-DDT is a poor insecticide, relative to *p,p'*-DDT, we suggest that consideration be given to decreasing the relative content of (±)-*o,p'*-DDT in the commercial technical grade DDT (possibly by a single crystallization from ethanol). This would increase the efficacy of commercial DDT as an insecticide and reduce the amount of (±)-*o,p'*-DDT, a known estrogen, being deployed in the biosphere.

EXPERIMENTAL SECTION

(±)-1-Chloro-2-[2,2,2-trichloro-1-(4-chlorophenyl)-ethyl]benzene (I). Technical grade DDT (9 kg) dissolved in 66 l. of hot ethanol was crystallized at room temperature and −15 °C yielding predominantly *p,p'*-DDT crystals. The mother liquors were evaporated and the residue dissolved in pentane, seeded with *p,p'*-DDT, and crystallized at room temperature and −15 °C. The crystals formed were predominantly *p,p'*-DDT. The reduced pentane mother liquors were crystallized at room temperature and −15 °C after seeding with *o,p'*-DDT (I). The resulting crystals were predominantly I and were crystallized three times from methanol. The resulting (±)-I (516.9 g, 30.8%) contained about 1.3% *p,p'*-DDT by weight (GLC analysis): mp 72–74 °C; GLC and TLC typical of *o,p'*-DDT (I); NMR ($CDCl_3$) δ 5.77 (s, 1 H, benzylic), 7.16–7.61 (m, 7 H, aromatic), 8.02–8.22 (m, 1 H, aromatic). Anal. Calcd for $C_{14}H_9Cl_5$: C, 47.4; H, 2.6; Cl, 50.0. Found: C, 47.8; H, 2.6; Cl, 49.7.

(±)-1-Chloro-2-[2,2,2-trichloro-1-(4-chlorophenyl)-ethyl]-4-nitrobenzene (II). (±)-I (50 g) was dissolved in 500 ml of CCl_4, the solution was cooled to 1 ± 1 °C, and 1.5 ml of concentrated H_2SO_4 was added. This solution was added to a 1 ± 1 °C solution of 112 ml of 90% HNO_3 in 500 ml of acetic anhydride previously mixed for 30 min at room temperature. The reaction, monitored on TLC, was stopped at about 2 h by pouring onto 3 l. of ice. NaOH (40%, 500 ml) was added and the aqueous phase extracted with pentane. The CCl_4 phase was evaporated and the residue dissolved in ethanol. The crude product was crystallized from ethanol twice and the pure product fractionally collected from an acid-washed alumina column eluted with 20% diethyl ether in hexane or petroleum ether (bp 38–47 °C) alone. The yield of pure (±)-NO_2-*o,p'*-DDT (II) recrystallized from ethanol was 22.1 g (39.3%); 14.3 g of I was recovered: mp 138–140 °C; NMR ($CDCl_3$) δ 5.78 (s, 1 H, benzylic), 7.26–7.67 (m, 5 H, aromatic), 8.17 (d of d, 1 H, J = 9.0, 3.0 Hz, aromatic), 9.09 (d, 1 H, J = 2.5 Hz, aromatic). Anal. Calcd for $C_{14}H_8Cl_5NO_2$: C, 42.1; H, 2.3; Cl, 44.4; N, 3.5. Found: C, 41.9; H, 2.1; Cl, 44.2; N, 3.3.

(±)-1-Chloro-2-[2,2,2-trichloro-1-(4-chlorophenyl)-ethyl]-4-aminobenzene (III). (±)-II (20 g), 20 g of mossy tin, and 70 ml of ethanol in a round-bottomed flask fitted with a condenser were placed under reflux for 10 min in a boiling water bath. HCl (100 ml) was added slowly and the mixture maintained under reflux with stirring for 1 h. The ethanol was removed by evaporation and 800 ml of H_2O, 800 ml of $CHCl_3$, and 200 ml of 40% NaOH were added and shaken. The $CHCl_3$ was removed, the solution twice extracted with diethyl ether, and the ethereal extract dried with Na_2SO_4, filtered, and used directly in salt formation. The amine for analysis was crystallized from petroleum ether (bp 38–47 °C): mp 108–111 °C; NMR ($CDCl_3$) δ 3.68 (s, 2 H, amine), 5.67 (s, 1 H, benzylic), 6.52 (d of d, 1 H, J = 8.5, 2.5 Hz, aromatic), 7.05–7.60 (m, 6 H, aromatic). Anal. Calcd for $C_{14}H_{10}Cl_5N$: C, 45.5; H, 2.7; Cl, 48.0; N, 3.8. Found: C, 45.5; H, 2.9; Cl, 48.0; N, 3.5.

(±)-1-Chloro-2-[2,2,2-trichloro-1-(4-chlorophenyl)-ethyl]-4-aminobenzene-(+)-7,7-dimethyl-2-oxobicyclo[2.2.1]heptane-1-methanesulfonate Salt. For the original resolution of the (−)-NH_2-*o,p'*-DDT (III) via fractional crystallization, 23.1 g (0.06 mol) of (±)-III was combined with 15.2 g (0.07 mol) of (+)-7,7-dimethyl-2-oxobicyclo[2.2.1]heptane-1-methanesulfonic acid ((+)-camphor-10-sulfonic acid) in CH_3CN. For the resolution of (−)-NH_2-*o,p'*-DDT and (+)-NH_2-*o,p'*-DDT by the reflux technique approximately 54.1 g (0.15 mol) of (±)-III in diethyl ether was combined with 34.0 g (0.15 mol) of (+)-camphor-10-sulfonic acid in CH_3CN, the solvents evaporated, and the salts (mp 177–183 °C dec) redissolved in 2 l. of CH_3CN.

(−)- and (+)-NH_2-*o,p'*-DDT-(+)-camphor-10-sulfonate salts. The reflux technique described in the text gave 12.4 g (14.1%) of (−)-NH_2-*o,p'*-DDT (III) salt, mp 297–298 °C dec, and 7.6 g (8.6%) of (+)-NH_2-*o,p'*-DDT (III) salt, mp 288–290 °C dec.

(−)- and (+)-NH_2-*o,p'*-DDT (III). (−)-Amine salt (11.0 g) was dissolved in a small volume of hot ethanol and cooled and an excess of 10% HCl was added. The resulting solution was stirred, heated to about 70 °C, cooled, made basic with Na_2CO_3, and extracted with diethyl ether. The extract was dried with Na_2SO_4, filtered, and evaporated. (+)-Amine salt (6.4 g) was treated similarly.

For the (−)-NH_2-*o,p'*-DDT, $[\alpha]^{25}D$ was −170.1° (*c* 0.55, ethanol) while for the (+)-NH_2-*o,p'*-DDT, $[\alpha]^{25}D$ was 166.4° (*c* 0.75, ethanol).

(−)- and (+)-*o,p'*-DDT (I). H_2SO_4 (200 ml, 12 N) and 400 ml of glacial acetic acid, heated to about 70 °C, were added to about 6.8 g of (−)-III. The amine–hydrogen sulfate dissolved and the solution was cooled to 1 ± 1 °C. $NaNO_2$ (1.83 g) in a small volume of water was added slowly with stirring and the solution stirred another 1 h at 1 ± 1 °C. H_3PO_2 (336 ml, 50%) was added, stirred 15 min, and left at 5 ± 2 °C for 72 h. H_2O (675 ml) plus 600 ml of 40% NaOH were added and the solution extracted with pentane. The pentane extract was evaporated and eluted from an acid-washed alumina column using petroleum ether (bp 38–47 °C). The (−)-*o,p'*-DDT (I) weighed 5.93 g (91.4%): mp 73.5–75 °C. Anal. Calcd for $C_{14}H_9Cl_5$: C, 47.4; H, 2.6; Cl, 50.0. Found: C, 47.1; H, 2.6; Cl, 50.3.

Similar ratios of reactants were used to deaminate the (+)-III. The (+)-*o,p'*-DDT (I) weighed 3.36 g (89.8%): mp 73–75 °C. Anal. Calcd for $C_{14}H_9Cl_5$: C, 47.4; H, 2.6; Cl, 50.0. Found: C, 47.5; H, 2.5; Cl, 50.5.

Both enantiomers were crystallized from ethanol and TLC, GLC, and NMR analyses for both were typical of racemic *o,p'*-DDT. The specific rotations were $[\alpha]^{25}D$ −17.9° (*c* 5.05, ethanol) and $[\alpha]^{25}D$ 17.7° (*c* 2.22, ethanol). The 1.3% *p,p'*-DDT contamination in the starting material was eliminated during the resolution process.

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Consistency of Toxaphene Composition Analyzed by Open Tubular Column Gas–Liquid Chromatography

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Each of 8 toxaphene samples manufactured by Hercules from 1949 to 1975 shows the same 29 major peaks in almost identical ratios based on analyses by open tubular column gas–liquid chromatography (GLC) with a linear electron capture detector. About 85% of the total peak area is accounted for by these 29 peaks which individually vary from 1 to 8% of the total. The 8 toxaphene samples from Hercules are easily differentiated by open tubular column GLC from 12 samples of related chlorinated terpenes from other manufacturers in the United States and abroad and from [^{14}C]- and [^{36}Cl]toxaphene prepared by Hercules. A more detailed analysis of toxaphene composition is provided by open tubular column GLC of toxaphene components in each of five TLC regions which are precisely defined by the use of selected fluorene marker dyes. Despite large composition differences between some of the samples, there is surprisingly little variation in their mouse intraperitoneal and housefly topical LD_{50} values.

Following the introduction of toxaphene by Hercules in the late 1940's (Buntin, 1951), several other companies in the United States and abroad have produced and marketed similar insecticides prepared by the chlorination of camphene and related terpenes. Food and feed containing residues of toxaphene and related materials are regulated on the basis of tolerances derived from analytical data using methods developed for toxaphene (Guyer et al., 1971; Zweig and Sherma, 1972) and from dietary no-effect levels in chronic feeding studies with toxaphene from Hercules (Lehman, 1965). These residue methods and toxicology data are only suitable for use with materials that closely approximate the composition of Hercules toxaphene. It is therefore important to intercompare the composition of toxaphene samples manufactured by Hercules over the past 26 years and of related commercial materials.

Toxaphene with an overall average molecular formula of $C_{10}H_{10}Cl_8$ is a complex mixture of at least 177 components revealed by a combination of liquid adsorption column chromatography followed by GLC–CIMS analysis on a packed column of the resulting fractions (Holmstead et al., 1974). An improved procedure for separation and quantitative analysis of toxaphene components is needed to critically intercompare the composition of toxaphene samples and related materials. This report gives an open tubular column GLC method for toxaphene analysis and applies this procedure to 8 samples of toxaphene manufactured by Hercules from 1949 until 1975, to 12 samples of toxaphene-like materials from other manufacturers, and to samples of [^{14}C]- and [^{36}Cl]toxaphene. It also evaluates a TLC–GLC method for more complete separation and analysis of toxaphene components and the effect of composition on the acute toxicity of toxaphene-like materials.

MATERIALS AND METHODS

Samples. Charles L. Dunn (Hercules Inc., Wilmington, Del.) provided the following samples: standard toxaphene and toxaphene batches manufactured by Hercules in 1949, 1954, 1957, 1960, 1963, 1970, and 1975; [^{14}C]toxaphene (1.35 mCi/g) and [^{36}Cl]toxaphene (43.6 μCi/g) prepared by Hercules; Hercasa product from the Hercules owned plant at Managua, Nicaragua. He also supplied additional samples believed to originate from the following sources: two samples from Vicksburg Chemical Co. (Vicksburg, Miss.); two samples from Bison Chemical Co. and one from Sonford Chemical Co. (both at Fort Natchez, Tex.); one sample from Procida (Paris, France). Strobane T-100 was supplied by Roy T. Gottesman (Tenneco Chemicals, Piscataway, N.J.). Kenneth R. Hill (Agricultural Environmental Quality Institute, United States Department of Agriculture, Beltsville, Md.) provided four samples: Flit & Fontaine manufactured in South Africa; Melipax

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