Figure 5. A. Salmon oil. B. Herring oil. Instrumental conditions given in text.

the rest. The degradation pattern for a mixture of p,p'-DDE and PCB peak 5 is presented in Figure 4.

Analysis of Salmon and Herring Oil. Figure 5 shows the chromatograms of salmon and herring oil extracts. The pattern for Aroclor 1254 was recognizable, but the ratio of the peak areas was not the same as the standard. Since the uv degradation of the PCB's is independent of the concentration, the degradation pattern will be the same for environmental samples as it is for standards unless preferential metabolism or degradation of some of the PCB's has occurred.

In analyzing salmon and herring oil, the peaks that apparently contained only PCB's yielded degradation patterns with the same peak area ratios as the standard. An example of this is given in Table III, salmon and herring oil peak 2 and PCB peak 4. When some other compound

was present, or perhaps preferential metabolism had occurred, the pattern was similar to the standard but the ratios were different. This can be seen by looking at the results in Table III for salmon and herring oil peak 4 and PCB peak 7.

Peak 3 of the oils was shown to contain PCB peaks 5 and 6 and p,p'-DDE. When the entire peak was trapped and irradiated, the degradation pattern could be used for the identification of only peak 6 and p,p'-DDE. By making two trappings of this peak, the first trap being removed at the top of the peak and replaced with a second trap, the presence of peak 5 was demonstrated.

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Mass Spectrometric Identification of the Hepta- and Octa-Chlorinated Dibenzo-p-dioxins and Dibenzofurans in Technical Pentachlorophenol

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Analytical measurements indicate that technical pentachlorophenol, a widely used pesticide, contained chlorinated dibenzo-p-dioxins. High-resolution mass spectrometry was used to determine the elemental composition of ions present in the spectrum of the mixture obtained as a cleaned-up neutral fraction from technical pentachlorophenol. Hepta- and octachlorodibenzo-p-dioxins were identified in the sample, as were hepta- and octachlorodibenzofurans.

Mass spectrometry is continuing to gain in importance as a method for determination of the structures of pesticides and their transformation products. Spectra can be obtained with isolated samples or with individual components of mixtures if a gas chromatograph is linked with

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the mass spectrometer through a suitable interface (Hutzinger *et al.*, 1971).

High-resolution mass spectrometry permits determination of the accurate masses of ions produced in the mass spectrometer; consequently the elemental composition of individual peaks in the spectrum can frequently be elucidated. The spectrum afforded by a mixture of compounds in the high-resolution mass spectrometer is complex, but potentially provides a method by which the individual components of a mixture can be recognized. Lovins (1969)

demonstrated that a mixture of pesticides can be analyzed by identification of ions in the mass spectrum of the mixture. If the ions identified are those of elemental composition uniquely defined for a particular pesticide, it necessarily follows from the appearance of such lines in the spectrum that the pesticide is a component of the mixture.

Although individual isomers cannot be distinguished, the technique may be useful for the identification of traces of biologically active materials. Baughman and Meselson (1971), for example, have described the potential use of high-resolution mass spectrometry for the analysis of tetrachlorodibenzo-p-dioxins. Since chlorinated dioxins have been implicated as agents responsible for the teratogenic effects of the herbicide 2,4,5-trichlorophenoxyacetic acid (2,4,5-T) and also for chick edema disease (Hearings before Subcommittee on Energy, 1970), their occurrence in chlorophenol derivatives has been the subject of many recent studies. Our interest in the presence of chlorinated dioxins as contaminants in technical pesticides prompted us to examine technical pentachlorophenol by this technique to identify neutral compounds present as impurities.

EXPERIMENTAL SECTION

Low-resolution spectra were obtained in the solids probe of a Perkin-Elmer GC 270 mass spectrometer (ionizing voltage 80 eV) at a probe temperature of 50°. High-resolution spectra were determined on a CEC 21-110B mass spectrometer, and the spectra were recorded on a photoplate. The sample was introduced through a direct probe, and spectra were recorded at intervals while the probe was heated from 135 to 216° (ionizing voltage 70 eV). Technical pentachlorophenol used was collected by the former Pesticide Regulation Division of the U. S. Department of Agriculture. Samples were analyzed for dioxin content by the method previously described (Woolson et al., 1972).

The quantity of chlorinated dibenzo-p-dioxins was estimated by electron-capture gas-liquid chromatography on a Perkin-Elmer Model 900 gas chromatograph fitted with glass columns (1.83 m × 4 mm i.d.) packed with 5% OV-225 on Chromosorb W and equipped with a ^{63}Ni electron-capture detector. The temperature was programmed from 230 to 260° at 4° per min and finally held at 260° until octachlorodibenzo-p-dioxin was detected. Isomeric chlorinated dioxins had closely related retention times, whereas groups of compounds of differing degrees of chlorination were more efficiently separated. Therefore, quantities of dioxins were estimated by summation of peaks with closely related retention times matching those of authentic standards. The values thus obtained represent the content of individual dioxins based on degree of chlorination, irrespective of isomers.

A sample of technical pentachlorophenol in methanol was treated with KOH to remove phenols following the procedure (A, B, and C) of Woolson et al. (1972). Evapo-ration of the hexane extract gave a white solid which was analyzed by mass spectrometry and gas chromatography.

RESULTS AND DISCUSSION

Pentachlorophenol has had a variety of pesticidal applications and has been used as a herbicide, defoliant, insecticide, and fungicide. A major use currently is as a wood preservative. Technical "pentachlorophenol" is manufactured by the chlorination of phenol or chlorophenols and may contain both tetra- and pentachlorophenols. Since vigorous thermal conditions may be employed in the manufacturing process, a variety of side reactions can occur to give condensed materials. These by-products may include chlorinated dibenzo-p-dioxins, and an analytical survey of the polychlorodibenzo-p-dioxin content of a number of pesticides and chlorophenols, including pentachlorophenol, was recently completed (Woolson et al., 1972). The presence of polychlorinated dibenzofurans in the neutral fraction was indicated in a preliminary examination by combined gas chromatography–mass spectrometry (gc-ms). This technique was valuable for the identification of tetra and lower chlorinated dibenzo-p-dioxins, but proved of limited value for examination of mixtures of polychlorinated compounds of high molecular weight and low volatility.

Recently, however, Firestone et al. (1972) analyzed eight pentachlorophenol samples by gas chromatography and found that hexa-, hepta-, and octachlorodibenzo-p-dioxins were present. Hexachlorodioxins were present in all samples. The combined gc-ms technique was used to confirm the presence of hexa- and heptachlorodibenzo-p-dioxins and to identify hexa- and heptachlorodibenzofurans in the extract. They also reported the presence of tetra-, penta-, and octachlorodibenzofurans in pentachlorophenol samples. Polychlorodiphenyl ethers containing from four to ten chlorine atoms were also identified in pentachlorophenol and its sodium salt.

Mass spectra are extremely valuable for characterization of the chlorinated dioxins (Buu-Hoi et al., 1972). The molecular ion is intense and often represents the base peak. The relative intensities of group of peaks above the molecular ion at M + 2, M + 4 ... , M + 2n due to increasing content of ^{37}Cl indicate the degree of chlorination of the molecule. The fragmentation patterns are simple and losses of Cl, (CO + Cl), 2 Cl, and 2 (CO + Cl) from the molecular ion occur. Doubly charged molecular ions are also observed. For example, the molecular ion of 2,3,7,8-tetrachlorodibenzo-p-dioxin, a typical member of this class (Figure 1), is at m/e 320 ($C_{12}H_4{}^{35}Cl_4O_2$) and the principal fragments {at m/e 285 ($C_{12}H_4{}^{35}Cl_3O_2$), [relative abundance (r.a.) 10%]; 257 ($C_{11}H_4{}^{35}Cl_3O$) (r.a. 34%); 250 ($C_{12}H_4{}^{35}Cl_2O_2$) (r.a. 5%); and 194 ($C_{10}H_4{}^{35}Cl_2$) (r.a. 18%)} represent losses of Cl, (CO + Cl), 2 Cl, and 2 (CO + Cl), respectively. The most abundant ion in the spectrum is at m/e 322 ($C_{12}H_4{}^{35}Cl_3{}^{37}ClO_2$) (base peak) (Crosby et al., 1971). A doubly charged ion is observed at m/e 160 (r.a. 13%).
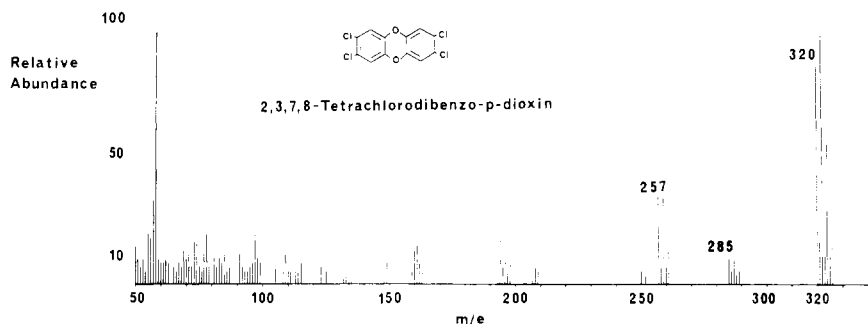

Figure 1. Mass spectrum of 2,3,7,8-tetrachlorodibenzo-p-dioxin.
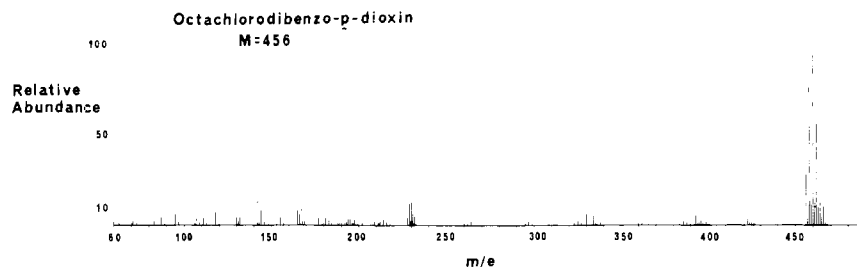
Figure 2. Mass spectrum of octachlorodibenzo-*p*-dioxin.

The unchlorinated parent compound, dibenzo-*p*-dioxin, loses CO or 2 CO from the molecular ion, whereas the chlorinated dioxins lose CO accompanied always by loss of Cl. The chlorinated compounds do not show any analogous path to the loss of a molecule of acetylene, which follows the loss of 2 CO molecules from dibenzo-*p*-dioxin (Calder *et al.*, 1970). The chlorinated dibenzofurans also possessed an intense molecular ion and lost a (CO + Cl) fragment.

The mass spectra of the higher polychlorinated dibenzo-*p*-dioxins resemble that of the tetrachloro compound. Octachlorodibenzo-*p*-dioxin has an intense molecular ion at m/e 456 (base peak at m/e 460) and a fragmentation pattern in which losses of 35 and 63 are observed (Figure 2). A doubly charged ion at m/e 228 is also a feature of the spectrum. Other features include intense ions at m/e 142 (C_6Cl_2) and 130 (C_5Cl_2).

For the examination of technical pentachlorophenol, a single commercial sample was used. It was anticipated that the spectrum of the crude neutral fraction obtained from pentachlorophenol might be very complex. However, the clean-up procedure requires a wash with 1:1 (v/v) HNO_3/H_2SO_4 mixture and this appears to remove many interfering impurities, although the recovery of some of the lower chlorinated dioxins may be reduced. Gas chromatographic analysis of the pentachlorophenol sample showed that the cleaned-up neutral fraction contained hexachlorodibenzo-*p*-dioxin (17 ppmw), heptachlorodibenzo-*p*-dioxins (108 ppmw), and octachlorodibenzo-*p*-dioxin (144 ppmw), identified by comparison of retention times with those of standard dioxins. The synthetic samples used contained unidentified isomers of four hexa and two heptachloro compounds. Unidentified peaks were present in the region where hexa- and heptachlorodi-

benzo-*p*-dioxins were eluted from the column. Evidence of the effectiveness of the clean-up procedure was obtained by a scan of the mass spectrum at unit resolution. The molecular ion and some fragment ions of hepta- and octachlorodibenzofurans, hepta- and octachlorodibenzo-*p*-dioxins could be clearly distinguished and were the major features of the spectrum. In addition, a group of peaks at m/e 376, 378, 380, etc., suggested the presence of a hexachlorodibenzofuran. The appearance of peaks at m/e 203, 211, 220, and 228 mass units and possessing relative abundances corresponding to the appropriate degree of chlorination can be explained by the presence of doubly charged ions corresponding to the molecular ions m/e 406, 422, 440, and 456 ($C_{12}H^{35}Cl_7O$; $C_{12}H^{35}Cl_7O_2$; $C_{12}{}^{35}Cl_8O$; and $C_{12}{}^{35}Cl_8O_2$), respectively. These are hepta- and octachlorodibenzofurans and dibenzo-*p*-dioxins.

The high-resolution spectra (Table I) provide unequivocal evidence for the allocation of empirical formulas, since each accurately measured ionic mass corresponds uniquely to that calculated for a particular combination of atoms. In a mixture of compounds the possibility that several components in the mixture might give ions of the same composition cannot be eliminated, and for unequivocal identification it is therefore necessary to choose a number of fragments characteristic of the compound under examination. Recognition of individual components in mixtures can be made by the definition of a limited number of characteristic fragment ions, but the analysis of complex mixtures by such a technique is preferably accomplished by a computer-based system.

The number of components of the mixture under examination was limited and the information obtained, particularly in the region of the molecular ions, gave clear evidence for the presence of octa- and heptachlorodibenzofurans and dibenzo-*p*-dioxins (Table I). Since the fragmentation pathways involve only losses of CO and/or Cl, there is little doubt that the ions at m/e 456, 440, 422, and 406 are truly molecular ions and that the accurate mass measurements 455.7400, 439.7452, 421.7782, and 405.7848 correspond to the calculated values 455.7406, 439.7457, 421.7796, and 405.7847 for the formulas $C_{12}Cl_8O_2$, $C_{12}Cl_8O$, $C_{12}HCl_7O_2$, and $C_{12}HCl_7O$, respectively (calcd for ^{12}C and ^{35}Cl). In addition, the masses of all isotopic peaks were calculated to show their agreement with measured values.

These findings provide confirmation of the presence of contaminant dioxins and dibenzofurans in some samples of technical pentachlorophenol, although it should be emphasized that the high-resolution spectra do not provide information concerning the orientation of the chlorine substituents in the heptachlorodibenzofurans and dibenzo-*p*-dioxins (Pohland *et al.*, 1971). Quantitative data previously published indicate that high levels of these contaminants may be present in some samples of technical pentachlorophenol; 6 of 20 samples contained more than 100 ppm of hepta- and octachlorodibenzo-*p*-dioxin (Woolson *et al.*, 1972). Although the pentachlorophenol sample under examination was manufactured before 1960, analysis of more recent samples reveals a similar dioxin

Table I. Calculated and Experimentally Determined Values of m/e for Sample of Neutral Pentachlorophenol Fraction.[a]
(All Isotopic Peaks Have Been Omitted but These Are in Agreement with Calculated m/e Values)

m/e found	m/e calcd for assumed composition	Composition	Standards
377	376.7826	376.7820	$C_{11}Cl_7$	$C_8F_{15}+$, $C_9F_{15}+$
386	385.8036	385.8029	$C_{12}Cl_6O_2$	
393	392.7735	392.7769	$C_{11}Cl_7O$	
405	404.7744	404.7769	$C_{12}Cl_7O$	$C_9F_{15}+$, $C_{10}F_{15}+$
406	405.7848	405.7847	$C_{12}HCl_7O$	
421	420.7691	420.7718	$C_{12}Cl_7O_2$	$C_8F_{17}+$, $C_9F_{17}+$
422	421.7782	421.7796	$C_{12}HCl_7O_2$	
440	439.7452	439.7457	$C_{12}Cl_8O$	$C_9F_{17}+$, $C_{10}F_{17}+$
456	455.7400	455.7406	$C_{12}Cl_8O_2$	$C_{11}F_{17}+$, $C_{12}F_{17}+$

[a] A complete table of m/e values (377–468) will appear following these pages in the microfilm edition of this volume of the journal. Single copies may be obtained from the Business Operations Office, Books and Journals Division, American Chemical Society, 1155 Sixteenth St., N.W., Washington, D. C. 20036. Remit check or money order for $6.00 for photocopy or $2.00 for microfiche, referring to code number JAFC-73-92.

content. For example, three samples collected in 1970 contained hexachlorodibenzo-p-dioxin (0.5 to 37 ppmw) and heptachlorodibenzo-p-dioxin (90 to 135 ppmw). In 1969 more than 45 million pounds of pentachlorophenol was produced in the United States and more than 25 million pounds of this was used as a wood preservative (Fowler et al., 1971). It would appear important, therefore, that investigation of the content and effects of chlorinated dibenzo-p-dioxins and chlorinated dibenzofurans in technical pentachlorophenol be continued.

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Comparison of Electron Capture and Electrolytic Conductivity Detectors for the Residue Analysis of s-Triazine Herbicides

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The capabilities of [63]Ni electron capture (ECD) and electrolytic conductivity detectors (CCD) have been compared for the analysis of s-triazine herbicides in water, soil, and corn samples by gas chromatography. Atrazine was selected for the residue study and other s-triazines were studied using various stationary phases only for their gas chromatographic characteristics. Quantitative recovery of atrazine from fortified water samples was obtained using dichloromethane. Extraction by acetonitrile, methanol, and acetone showed good recovery for soil samples. Acetonitrile extraction was found adequate for corn analysis. The cleanup procedures involved partitioning and column chromatography on deactivated alumina. Comparable sensitivities were obtained using both methods of detection and quantitation for the range studied (0.02 to 2.0 ppm). The CCD seemed to have wider application than the ECD, which required cleanup in all cases studied. The conductivity analysis of water and soil samples could be performed quantitatively without cleanup, whereas corn samples required cleanup.

The chemical analysis of triazine herbicides in soils has been reviewed by Mattson et al. (1970); they gave the detailed method for the analysis of atrazine in soil samples on a routine basis using acetonitrile-water-mixed solvent extraction, cleanup by alumina column, and microcoulometric gas chromatography.

The use of gas chromatography for the analysis of triazine herbicides in various substrates has also been reviewed recently (Cochrane and Purkayastha, 1972). In this review the different aspects such as extraction, cleanup, gas chromatographic separation, and detectors for the analysis of various herbicides have been presented.

These two reviews state that the following solvents have been used for the extraction of triazines: dichloromethane, chloroform, and diethyl ether for water; chloroform, di-

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chloromethane, methanol, acetone, and acetonitrile-water for soil; and methanol(ammoniacal)-dichloromethane, methanol, chloroform, aqueous acetone, Skellysolve B, and acetonitrile for plant samples.

The water samples needed minimum or no cleanup. Partition and chromatography on alumina columns have been used for the removal of impurities from soil samples. But for plant samples, a further cleanup on a Florisil column has been found necessary in a few instances.

Recently the selectivity and sensitivity of the Coulson electrolytic conductivity detector (CCD) (Coulson, 1966) for nitrogen-containing pesticides have been shown to be satisfactory (Cochrane and Wilson, 1971; Eberle and Hörmann, 1971; Hormann and Eberle, 1971; Patchett, 1970; Ramsteiner et al., 1971; Westlake et al., 1970). Also, analysis of atrazine residues in a field soil and comparison of several extraction methods using an electron capture detector (ECD) have been reported (Purkayastha, 1971).

The purpose of the present work was to compare the capabilities of ECD and CCD for the residue analysis of s-triazine herbicides in water, soil, and corn samples by