Journal of Chromatography, 303 (1984) 351–360
Elsevier Science Publishers B.V., Amsterdam — Printed in The Netherlands

CHROM. 17,019

STUDY OF CHLORINATED DIPHENYL ETHERS AND CHLORINATED 2-PHENOXYPHENOLS AS INTERFERENCES IN THE DETERMINATION OF CHLORINATED DIBENZO-*p*-DIOXINS AND CHLORINATED DIBENZOFURANS IN BIOLOGICAL SAMPLES

JOHN J. RYAN*, RAYMONDE LIZOTTE and WILLIAM H. NEWSOME
Food Research Division, Health Protection Branch, Tunney's Pasture, Ottawa, Ontario K1A 0L2 (Canada)
(Received May 21st, 1984)

SUMMARY

Two classes of environmentally occurring chlorinated aromatic hydrocarbons which have mass spectral properties similar to the chlorinated dibenzo-*p*-dioxins (dioxins) and chlorinated dibenzofurans (furans) were studied. Standards of chlorinated diphenyl ethers (CDEs), and chlorinated 2-phenoxyphenols (CPPs) and their methyl ethers, along with the dioxins and furans were passed in steps through a simple method for the analysis of the latter compounds in biological samples. The CDEs, which interfere with the determination of furans by mass spectrometry, had similar extraction, high-performance liquid chromatographic and gas chromatographic properties as the furans but, in all cases studied, were well separated from them on an activated Florisil column using combinations of hexane and dichloromethane as eluting solvents. The higher CPPs on the other hand tended to generate dioxin residues by ring closure when exposed to strong hydrochloric acid solution during sample preparation. In addition, their methyl ethers containing four to six chlorines tracked completely through all stages of the method with the dioxins. Thus, if the methyl ethers of CPPs were present in a sample extract, additional mass spectral information would be needed to further differentiate them from dioxins in environmental samples.

INTRODUCTION

Chlorinated dibenzo-*p*-dioxins (dioxins) and chlorinated dibenzofurans (furans) are two classes of environmental contaminants which have attracted a great deal of study in the last decade because of their potent toxic properties. As a result, many methods[1] have been developed for their determination in biological samples the majority of which use gas chromatography (GC)–mass spectrometry (MS) in the measurement procedure. These methods have to distinguish dioxins and furans in sample extracts from other classes of organic compounds which have similar MS properties and occur together in environmental and biological samples. In particular, two classes which have similar MS fragmentation are the chlorinated 2-phenoxyphenols (CPPs)

0021-9673/84/$03.00

and their methyl ethers for the dioxins and the chlorinated diphenyl ethers (CDEs) for the furans.

The CPPs and their methyl ethers readily generate dioxins by the loss of either HCl or CH_3Cl. This transformation is known to occur with heat[2] or photolysis[3] (hence the common name, pre-dioxin) and also takes place in the mass spectrometer[4,5] to give an ion cluster fragment of exactly the same mass as a dioxin with one chlorine less. The second class of interfering compounds, the CDEs, form furans both photochemically[6] and thermally[7]. Moreover during MS, they fragment by the loss of two chlorines to give the exact mass as a furan of two chlorines less[8,9]. Thus any analytical technique for the measurement of dioxins and furans must distinguish them from the CPPs and CDEs all of which are known to occur together[8–10] in chlorophenol preparations.

Previous studies on the CPPs[8–11] have shown their extraction into organic solvents under neutral conditions and their separation from dioxins by adsorption on alumina and Florisil. No work has been carried out on the methylated CPPs which could arise in environmental samples by biological methylation in analogy to the formation of chlorinated anisoles from chlorinated phenols in chicken products[12]. Some work has been reported on the properties of CDEs on alumina[8,9] and Florisil[13] columns but their chromatographic relationship vis-à-vis furans is not clear. Smith and Johnson[14] investigated a series of potentially interfering chlorinated aromatic hydrocarbons in a specific method for dioxin and furan determination. However, no separation of the method into its component parts and a study of the individual steps was carried out.

The purpose of this work was to study the chromatographic and separation properties of a series of specific standards of CPPs and CDEs as applied in steps to the method currently used in our laboratories for the determination of dioxins and furans in biological samples. It is shown that the CPPs and CDEs are readily separated by our method from the dioxins and furans, respectively, before the MS determinative step. However, some of the CPPs can be inadvertently converted to dioxins during the sample preparation steps and their methyl ethers behave very similarly to the dioxins throughout all stages of the method.

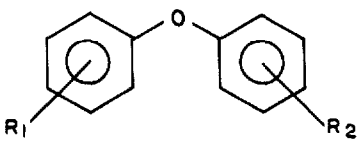
EXPERIMENTAL

Chlorinated aromatic hydrocarbon standards

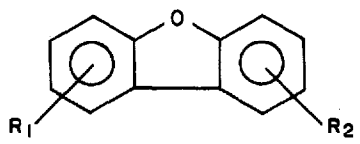
The classes of compounds studied are illustrated in Fig. 1. The CDEs were synthesized as described previously[13]. The dibenzofuran standards with 2,3,7,8-chlorosubstitution were either synthesized from the corresponding ether using palladium acetate dehydrogenation[15] or obtained as gifts. Some of the CPP standards were synthesized and methylated as described[11] and others were obtained by donation. The dioxins were obtained either from commercial sources or by exchange with other research groups. All standards had adsorption chromatographic, GC and MS properties in accordance with their structures or as described in the literature.

Florisil column chromatography

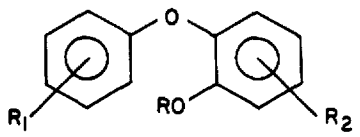
All glassware was rinsed twice with those solvents in which it was subsequently in contact. Florisil, 60–100 mesh, was obtained from the Floridin Co., (Berkeley



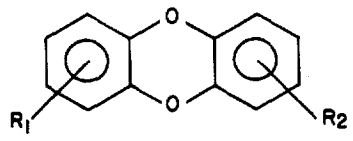
Chlorinated Diphenyl Ethers
(CDE)
R_1, R_2 = H, Cl



Chlorinated Dibenzofurans
R_1, R_2 = H, Cl



Chlorinated 2-Phenoxyphenols
(CPP)
R = H, CH_3
R_1, R_2 = H, Cl



Chlorinated Dibenzo-p-dioxins
R_1,R_2 = H, Cl

Fig. 1. Chemical structures of chlorinated aromatic hydrocarbons studied in this report.

Springs, WV, U.S.A.). It was Soxhlet extracted in about 500-g portions with 3-l portions of dichloromethane until a 300:1 concentration of eluting solvent showed a flat baseline (less than 10% full scale deflection of any peak) at attenuation 32 on an OV-17 packed column at 220°C with electron-capture detection (ECD). The gravity columns used were disposable borosilicate glass serological graduated pipettes (Fisher Scientific) of nominal volume 5 ml, 27 cm × 6 mm I.D. A plug of dichloromethane-washed glass wool was inserted into the tapered end and the column filled with tapping with 1.5 g of the purified Florisil up to about the 2-ml mark. A 1-cm layer of dichloromethane-washed sodium sulfate was added and, finally, a plug of glass wool. The adsorbant was activated by heating the prepared column at 130°C for 24 h in an oven. After activation, the columns were cooled for 30 min and used within 90 min. They were first washed with 10 ml of dichloromethane and 10 ml of hexane–dichloromethane (98:2, v/v) and the washings discarded. The sample extract or standard was applied to the column with a Pasteur pipette in 1–2 ml of hexane. The column was then eluted with 20 ml of hexane–dichloromethane (98:2) which was collected in a 50-ml round bottomed flask. This fraction is known to contain polychlorinated biphenyls (PCBs) and, while it is ordinarily discarded, in this study it was collected. The Florisil column was then eluted with a total of 35 ml dichloromethane at a head pressure to give an elution rate of about 1.5 ml/min and this fraction was also collected. The solvents in both fractions were evaporated to a small volume on a rotary evaporator under reduced pressure using a room temperature water bath and then transferred in portions to a 0.3-ml conical vial with a Pasteur pipette. After each addition, the solvents were evaporated to a small volume and finally just to dryness with the aid of a jet of purified nitrogen and the residue taken up in a small

(25–100 μl) volume of toluene or isooctane for GC analysis, or acetonitrile for high-performance liquid chromatography (HPLC).

HPLC

This was carried out using the C_{18} reversed-phase system with methanol as the eluent as described earlier[16,17].

GC

Several types of GC instrumentation and detectors were used. For most recovery studies, a Hewlett-Packard Model 5700A gas chromatograph was equipped with a ^{63}Ni electron-capture detector and fitted with a 120 cm × 4 mm I.D. glass column containing 1% OV-17 on Chromosorb W with argon–methane (95:5) at a flow-rate of 60 ml/min. Column temperatures between 190 and 250°C were chosen to give retention times between 4 and 10 min. Some difficult separations were carried out on a Varian 1400 gas chromatograph equipped with a scandium tritide electron-capture detector and fitted with a 10 m × 0.25 mm I.D. fused-silica OV-17 capillary column. The splitless injector was maintained at 250°C, the helium carrier gas at a linear flow-rate of 40 cm/sec, and the nitrogen electron-capture detector make-up gas at 20 ml/min. Isothermal column temperatures were chosen between 190 and 240°C to give retention times between 7 and 25 min. Some samples were also confirmed by the newly developed technique of rapid chromatography–tandem MS consisting of a Hewlett-Packard 5840 gas chromatograph interfaced to a TAGA 6000 triple quadrupole mass spectrometer[18,19]. The GC column was a wide-bore (0.32 mm) 12 m bonded phase DB-5 fused-silica capillary column with nitrogen the carrier gas at 1.8 ml/min. An aliquot of sample was cold trap injected at 100°C, the gas chromatograph linearly programmed at 30°C/min to 160°C, then at 15°C/min to 280°C. The tandem MS system consisted of a Townsend low pressure chemical ionization source using purified air (80% nitrogen, 20% oxygen) as reagent gas. The positive ions generated were selected by the first quadrupole (Q_1) and subsequently fragmented by collision induced dissociation with argon gas in the second quadrupole (Q_2) region. The specific fragmentation ions (daughter ions) after the loss of COCl were separated and analysed by the third quadrupole (Q_3). Both GC temperature and MS multi-reaction monitoring parameters were chosen to give a total time of about 20 min between injections.

RESULTS

The method used in our laboratories for the determination of dioxins and furans in biological matrices is a four- or five-step procedure depending on sample type, and requirements for specificity and sensitivity. The homogenized sample or aliquot is first extracted with acetone–hexane (2:1, v/v)[20] and lipid in the hexane portion is degraded and removed with concentrated sulphuric acid. The organic extract is next passed through an activated Florisil column which is known to separate PCBs from dioxins and furans. In some cases further clean-up or confirmation is carried out by reversed-phase HPLC fractionation on a C_{18} column. Lastly, determination is effected by GC–MS with both polar and non-polar columns and with low and high-resolution magnetic MS, and tandem MS being used. For this reason

the method was broken down into its component parts and, for each part, the behaviour of CPPs and CDEs with respect to dioxins and furans investigated.

Chlorinated diphenyl ethers

Florisil. To investigate the effect of Florisil column chromatography on the CDEs, five sets of CDE–furan standards were run in duplicate together through columns at spiking levels between 2 and 5 ng and the distribution between the hexane–dichloromethane (98:2) and dichloromethane fractions monitored by GC–ECD. The furans chosen were those isomers with four or more chlorines and a 2378-substitution pattern on the aromatic ring (biologically important isomers) as listed in Table I. For each furan a corresponding CDE was chosen, having two additional chlorines and separable from the furan by GC. Thus such a CDE would generate a positive fragment in the MS at the exact molecular ion of a furan of two chlorines less. The relative retention times (RRT) (Table I) on OV-17 of the CDEs with respect to the corresponding 2378-substituted furans vary between 0.7 and 1.6, a range for which one class could easily be confused with another, particularly when 210 and 135 congeners are known for the CDE and furans, respectively. Average recoveries of the CDEs and furans from the activated Florisil columns are given in Table I. These results show that the CDEs are eluted in the first fraction and the furans in the second fraction in every case irrespective of the degree of chlorination or the substitution pattern and that the recoveries are acceptable. As a further check on the capacity and separation powers of the Florisil adsorbant, the 23478-substituted furan and 234-2356-substituted CDE were run in duplicate together through Florisil columns at a loading of 5 ng for the furan and 500 ng for the CDE. In this instance, less than 0.05% (0.25 ng) of CDE was found in the dichloromethane fraction while the furan recovery was the same as in the equal loading case. Even if

TABLE I

RECOVERIES FROM ACTIVATED FLORISIL COLUMNS AND RELATIVE RETENTION TIMES (RRT) OF FIVE SETS OF CDE AND 2378-SUBSTITUTED CHLORINATED FURAN STANDARDS

Chlorine substitution		*Recoveries (%)**				*RRT***	
CDE	*Furan*	*Hexane–dichloromethane (98:2)*		*Dichloromethane*		*GC****	*HPLC****
		CDE	*Furan*	*CDE*	*Furan*		
234-345	2378	73	–§	–	103	1.55	0.95
234-2356	23478	88	–	–	100	1.21	0.74
2345-2345	123678	113	–	–	114	1.19	0.58
2345-23456	1234678	89	–	–	80	1.09	0.57
23456-23456	12346789	84	–	–	87	0.67	0.60

* Average values of duplicate determinations of 2–5 ng fortifying levels as determined by GC–ECD.

** CDE Relative to the 2,3,7,8-chlorinated furan of two chlorines less.

*** OV-17 Packed column for GC and reversed-phase C_{18} column with methanol as eluent for HPLC.

§ Dash means less than 1% of either CDE or furan appeared in this fraction.

the CDE were present in a 100-fold excess as might occur in certain samples, the Florisil was capable of discriminating between the two. Thus, even though there is a similarity in chemical structure and polarity, CDEs can be readily separated from furans on an activated Florisil column.

HPLC and GC. GC and HPLC properties of some CDEs with respect to the furans are listed in Tables I and II. These data show generally that, for the same chlorine numbers (Table II), CDEs are eluted earlier in both GC and reversed-phase HPLC than furans. CDEs with two chlorines more than the furan (Table I) are eluted in GC in most cases later than the furan and in HPLC earlier but near the furan. Thus, when a wide HPLC fraction is taken or when a gas chromatogram with many peaks results, confusion between classes would be possible and even probable.

TABLE II

RELATIVE RETENTION TIMES (RRT) OF CDE RELATIVE TO FURANS WITH 2378-SUBSTITUTION

Chlorine substitution		*RRT**	
CDE	*Furan*	*GC***	*HPLC****
34-34	2378	0.51	0.69
234-34	23478	0.54	0.60
345-34	12378	0.72	0.49
234-345	123678	0.53	0.43
234-234	234678	0.58	0.50
2345-234	1234678	0.61	0.36
2345-345	1234789	0.34	0.38
2345-2345	12346789	0.48	0.33

* CDE Relative to 2,3,7,8-chlorine substituted furan.
** 1% OV-17 Packed column at column temperatures between 200 and 240°C.
*** Reversed-phase C_{18} column with 100% methanol as eluent.

Chlorinated phenoxy phenols and their methyl ethers

Sulfuric acid treatment. The effect of strong acids on CPPs was investigated by fortifying solutions with 10-μg quantities of various CPPs and subjecting them to either sulfuric acid partition[16] or by shaking them with concentrated hydrochloric acid, a common technique[21] for the dissolution of connective and other tissues in dioxin and furan analysis. The extracts were then processed through the Florisil column clean-up and analyzed by both GC–ECD and verified by GC–MS. The results in Table III show that only 2-(2,3,4,5,6-pentachlorophenoxy)-3,4,5,6-tetrachlorophenol (nonachloro-2-phenoxyphenol) is converted in small amounts to the corresponding dioxin by strong acid and then only by hydrochloric and not by sulfuric acid. The heptachloro- and pentachloro-2-phenoxyphenols were not converted to either the tetra- or hexachloro-dibenzodioxin, respectively, and appear stable under these conditions. Even this small percentage conversion of nonachloro-2-phenoxyphenol to OCDD would represent a residue level in the order of 1 ppb* on a 10-g tissue sample which originally contained 1 ppm of CPP. The conversion of the higher chlorinated 2-phenoxyphenols to the corresponding dioxin by acid is similar to their

* The American billion (10^9) is meant.

TABLE III

EFFECT OF ACID TREATMENT ON THE CONVERSION OF CPP TO THE CORRESPONDING DIOXIN

Chlorine substitution pattern of (2-phenoxy)-phenol	*Equivalent dioxin*	*Conversion* (%)*		
		Control, no acid	*Conc.** hydrochloric acid*	*Conc.*** sulfuric acid*
23456-0	1234	<0.05	<0.05	<0.05
23456-45	123478	<0.01	<0.01	<0.01
23456-3456	12346789	<0.01	0.086	<0.01

* Mean of duplicate determination of 10 μg fortification.
** Methanol solution (1 ml) shaken with 60 ml acid for 2 h.
*** Methanol (1 ml) and 60 ml hexane shaken with 10 ml acid for 1 h.

conversion by heat whereby the lower chlorinated analogues do not lose HCl even at 250°C as occurs in a GC injection port while the higher CPPs do in fact ring close to the dioxin[22].

Florisil. CPPs are known to be strongly adsorbed on Florisil and only eluted by polar solvents such as methanol and ethyl acetate[11] and thus would not ordinarily be eluted in the dioxin fraction used in our method. Their methyl ethers are less polar as a result of which their elution pattern on Florisil was studied. Two sets of CPPs were made up, methylated with diazomethane, and their elution pattern on Florisil studied. As shown in Table IV, the methyl penta-, hexa- and heptachlorinated-2-phenoxyphenols are eluted completely in the dichloromethane portion and thus do not separate from the dioxin fraction. The methyl octa-CPP is split between the two elution fractions and the nona-CPP is eluted completely in the hexane wash and thus would be completely separated from the dioxins.

TABLE IV

RECOVERY* (%) OF METHYL CCP ON FLORISIL

Substitution pattern of (2-phenoxy)-phenol	*Equivalent dioxin*	*Recovery (%)*			
		Hexane–dichloro-methane (98:2) 1st 10 ml	*Hexane–dichloro-methane (98:2) 2nd 10 ml*	*Dichloro-methane, 35 ml*	*Total recovery*
23456-0	1234	–**	–	98	98
2356-45	12478	–	–	95	95
23456-45	123478	–	–	98	98
2345-345	123678	–	–	107	107
2345-3456	1234678	–	70	22	92
23456-3456	12346789	25	58	–	83

* Average of duplicate determinations of 5-ng portions.
** Dash means no methyl CPP detected in this fraction.

GC and HPLC. The CPPs are much more polar than dioxins. By GC they would not be eluted with the dioxins. Similarly, the CPPs in reversed-phase HPLC need water and acid for reproducible chromatography and, in a pure methanol system, are eluted with the solvent front. The methyl CPPs, however, are much nearer to the dioxins in their chromatographic behavior. This is illustrated in Table V where the relative GC and HPLC properties are listed for a series of methyl CPPs and the dioxin to which it is readily converted (loss of methyl chloride). For GC, the methyl CPPs are eluted both after (penta-, hexa-, and hepta-) and before (octa- and nona-) the equivalent dioxin *i.e.* their elution pattern in GC is narrower than that of the dioxins. For HPLC, the methyl CPPs are eluted earlier than the equivalent dioxin in every case but still within the same chromatographic region. The reversed-phase HPLC properties of these two classes are further illustrated in Fig. 2 showing clearly the earlier eluted methyl CPPs and the overlap between the two classes.

TABLE V

RETENTION TIMES (RRT) OF METHYL CPP RELATIVE TO THE EQUIVALENT DIOXIN (LOSS METHYL CHLORIDE)

Chlorine substitution pattern of (2-phenoxy)-phenol	*Dioxin equivalent*	*RRT*	
		*GC**	*HPLC***
23456-0	1234	1.23	0.54
2356-45	12478	1.13	0.56
23456-45	123478	1.03	0.58
2345-345	123678	1.30	0.59
2345-3456	1234678	0.77	0.63
23456-3456	12346789	0.72	0.66

* 1% OV-17 Packed column at column temperatures between 200 and 240°C.
** Reversed-phase C_{18} column with 100% methanol as eluent.

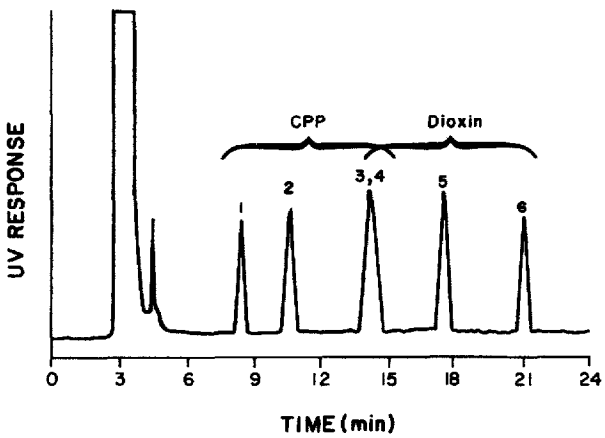


Fig. 2. Reversed-phase HPLC chromatogram with methanol as eluent of methyl CPPs and their chlorinated dibenzo-*p*-dioxin (dioxin) equivalent; elution conditions as in Experimental. Peaks: 1 = 2345-345-CPP; 2 = 2345-3456-CPP; 3 = 23456-3456-CCP; 4 = 123678-dioxin; 5 = 1234678-dioxin; 6 = 12346789-dioxin.

DISCUSSION

This study has taken a specific method for the determination of dioxins and furans in biological samples, and shown the effect of the method on two classes of organic compounds which interfere in the mass spectral determinative step and which occur in environmental samples. CDEs, furans, CPPs and dioxins are all known to be extracted from solid and liquid samples into acetone–hexane mixtures[11,13,18]. The effect of concentrated sulfuric acid on CDEs and methyl CPPs was not investigated. However, dioxins and furans containing four or more chlorines are known to be inert to this reagent on partitioning in hexane at room temperature and it is probable that similarly chlorinated CDEs and methyl CPPs exhibit the same inertness. Lower chlorinated (less than four) congeners of dioxins and furans are known to be partially sulfonated by shaking hexane solutions with concentrated sulfuric acid at room temperature and it is also possible that CDEs and methyl CPPs of less than four chlorines exhibit a similar behavior. However, none of these possibilities was specifically investigated.

With respect to the CDEs, the results show that they have similar GC, HPLC and extraction properties as the furans. However, CDEs are well resolved from the furans on Florisil columns with hexane and dichloromethane as eluting solvents, the amount or substitution of chlorination does not appear to be important, and a 100-fold excess of CDE over furan does not cause a change in the elution pattern. Thus in quantities that are likely to be present in biological samples the method will separate the CDEs from furans such that interference will not be a problem at the GC–MS determinative step. This result is in agreement with the work of Buser[9] which showed that alumina and the same elution solvents would separate these classes of compounds even though other solvents as diethyl ether and hexane[8] would not.

The situation with regard to the CPPs is more complicated. First of all there is now evidence that the higher chlorinated analogues will lose the elements of HCl when exposed to concentrated hydrochloric acid. As a result, a common laboratory procedure for sample preparation could conceivably generate a dioxin residue even though the precursor itself has such different polar and chromatographic properties that it is easily removed by subsequent separation techniques. This result adds to the known effects of heat[2] and light[3] as agents which generate dioxins from precursor CPPs. Similarly, the CPPs, as their methylated derivatives, have very similar extraction, GC, HPLC and Florisil elution properties as the dioxins particularly the lower chlorinated compounds. Any sample containing methyl CPPs could be confused in MS for a dioxin if only GC–MS monitoring of molecular ion clusters were carried out even if the sample and its extracts passed through a fairly rigorous clean-up procedure. Additional ions in the mass spectrometer as well as other GC column loadings would have to be assayed if methyl CPPs were suspected of being present in the sample extract. As such, this study is the first to demonstrate the real potential for methyl CPPs to interfere in dioxin analysis.

While methods for dioxin and furan measurement vary in many details, the basic components of extraction, partition, chromatography and detection are similar. Hence, other laboratories using different methods should be able to judge whether CDEs and CPPs will cause problems in their determination of furans and dioxins since we have broken our method into its component parts and studied the CDE and CPP behavior in each part.

ACKNOWLEDGEMENTS

The authors thank T. Deinzer of Oregon State University (Corvallis, OR, U.S.A.) and D. Firestone (U.S. Food and Drug Administration) for their generous gifts of some chlorinated phenoxyphenol, dioxin and furan standards.

T. Sakuma (Sciex Inc., Toronto, Canada) and B. Lau (Health Protection Branch, Ottawa, Canada) carried out the mass spectral determination in a precise and rapid manner.

REFERENCES

1 *Polychlorinated Dibenzo-p-dioxins: Limitations to the Current Analytical Techniques*, Publication NRCC 18576, National Research Council Canada, 1981.
2 C. Rappe and C.-A. Nilsson, *J. Chromatogr.*, 67 (1972) 247.
3 C.-A. Nilsson, K. Andersson, C. Rappe and S.-O. Westermark, *J. Chromatogr.*, 96 (1974) 137.
4 S. Jensen and L. Renberg, *Ambio*, 1 (1972) 62.
5 M. Deinzer, J. Lamberton, D. Griffin and T. Miller, *Biomed. Mass Spectrom.*, 5 (1978) 566.
6 A. Norstrom, K. Andersson and C. Rappe, *Chemosphere*, 5 (1976) 21.
7 R. Lindahl and C. Rappe, *Chemosphere*, 9 (1980) 351.
8 D. Firestone, J. Ress, N. L. Brown, R. P. Barron and J. N. Damico, *J. Ass. Offic. Anal. Chem.*, 55 (1972) 85.
9 H.-R. Buser, *J. Chromatogr.*, 107 (1975) 295.
10 C.-A. Nilsson and L. Renberg, *J. Chromatogr.*, 89 (1979) 325.
11 W. H. Newsome and J. B. Shields, *Intern. J. Environ. Anal. Chem.*, 14 (1983) 299.
12 R. F. Curtis, D. G. Land, N. M. Griffiths, M. Gee, D. Robinson, J. L. Peel, C. Dennis and J. M. Gee, *Nature (London)*, 235 (1972) 223.
13 W. H. Newsome and J. B. Shields, *J. Chromatogr.*, 247 (1982) 171.
14 L. M. Smith and J. L. Johnson, in G. Choudhary, L. H. Keith and C. Rappe (Editors), *Chlorinated Dioxins and Dibenzofurans in the Total Environment, Part 1*, Butterworth, London, 1983, p. 321.
15 A. Norstrom, K. Andersson and C. Rappe, *Chemosphere*, 6 (1976) 419.
16 J. J. Ryan and J. C. Pilon, *J. Chromatogr.*, 197 (1980) 171.
17 J. J. Ryan and J. C. Pilon, *J. Chromatogr.*, 248 (1982) 409.
18 J. J. Ryan, D. T. Williams, B. P.-Y. Lau and T. Sakuma, in L. H. Keith, G. Choudhary and C. Rappe (Editors), *Chlorinated Dioxins and Dibenzofurans in the Total Environment, Part 2*, Butterworth, London, 1984, pp. 205–214.
19 B. P.-Y. Lau, J. J. Ryan and T. Sakuma, 1984, in preparation.
20 R. Van Renterghem and L. Devlaminck, *Z. Lebensm. Unters. Forsch.*, 170 (1980) 346.
21 M. Langhorst and L. A. Shadoff, *Anal. Chem.*, 52 (1980) 2037.
22 J. J. Ryan, unpublished results.
23 J. J. Ryan, J. C. Pilon, H. B. S. Conacher and D. Firestone, *J. Ass. Offic. Anal. Chem.*, 66 (1983) 700.