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Polychlorinated Dibenzofurans (PCDFs)

Environmental Occurrence and Physical, Chemical and Biological Properties

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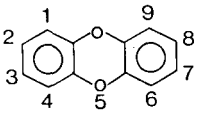
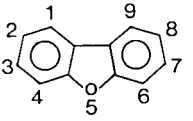
1. INTRODUCTION

Although much information is available on the fate of polychlorinated dibenzo-*p*-dioxins in the biotic and abiotic environment, in particular about the fate of 2,3,7,8-TCDD, information about polychlorinated dibenzofurans (PCDFs) is limited, despite the fact that large amounts of commercial compounds which contain PCDFs as contaminants are produced and are subsequently released into the environment. PCDFs are important, because their chemical and biological properties can closely resemble those of PCDDs. Such resemblance with PCDDs should create serious environmental concern, as large amounts of products containing PCDFs have already entered the environment.

This review contains currently available information about the physical, chemical and biological properties of PCDFs which could be useful in environmental studies. Methods for the chemical analysis of PCDFs have been excluded, as these procedures apparently correspond to those applied for PCDDs. The structures, numbering

and possible numbers of PCDF and PCDD isomers are shown in Figure 1.





Isomer groups	PCDD	PCDF
mono-	2	4
di-	10	16
tri-	14	28
tetra	22	38
penta-	14	28
hexa-	10	16
hepta-	2	4
octa-	1	1
	75	135

FIGURE 1 Structures, numbering and possible number of isomers of PCDD and PCDF.

2. PHYSICAL PROPERTIES

Available information about the physical properties of PCDFs indicate that PCDFs resemble PCDDs in behaviour. Data on melting point, vapor pressure, molar refraction, vapor density and rate of evaporation are comparable to those of PCDDs. The evaporation rate and vapor pressure/density decreases with an increasing number of chlorine atoms, while the melting point increases. Because of the low evaporation rate, direct atmospheric distribution of PCDFs from soil is of low environmental concern. In fact, most of the PCDF-distribution occurs when these compounds are adsorbed on particles. Although there are no data on adsorbtion and desorbtion on soil particles, environmental transport of PCDFs by leaching, for example, is likely to resemble that of PCDDs. Although the solubility of PCDFs in polar and non-polar solvents is currently not known, it can be expected to be of the same magnitude as found for PCDDs. Some physical properties of PCDFs are listed in Tables I and II (Firestone and U.S.E.P.A.[1,2]).

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TABLE I
Physical properties of some PCDF isomers (in ref. 2)

Compound	MP, °C	Vapor pressure (est.) 25°C	Molar refraction	UV max ($CHCl_3$), nm
DCDF			60.2	
2,4		7.3×10^{-6}		
3,7		7.0×10^{-6}		
2,8	185	6.8×10^{-6}		
TrCDF			65.0	
2,4,6	116–117	4.0×10^{-6}		
2,3,8	189–191	3.7×10^{-6}		256,302,313
2,4,7				
2,4,8				
TCDF			69.8	
1,4,6,8		2.5×10^{-6}		
2,4,6,8	198–200	2.5×10^{-6}		257,294,310,323
2,3,6,8		2.2×10^{-6}		
2,4,6,7		2.1×10^{-6}		
1,2,7,8		2.0×10^{-6}		
2.3.7.8	227–228	2.0×10^{-6}		259,309,316
2,3,6,7		1.9×10^{-6}		
3,4,6,7		1.8×10^{-6}		
PnCDF			74.6	
1,3,4,7,8		1.3×10^{-6}		263,272,297,320
1,2,4,7,8	234–235	1.3×10^{-6}		256,266,297
1,2,3,6,7		1.1×10^{-6}		
2,3,4,7,8		1.1×10^{-6}		
HpCDF			84.2	
		4.4×10^{-7}		
		3.6×10^{-7}		
		3.0×10^{-7}		
OCDF		1.9×10^{-7}	89.0	

TABLE II
Vapor density[a] and evaporation rate[b] of some PCDF isomers. (In ref. 2)

Compound	Vapor density (g/cm^3)	$Q(g/cm^2/s)$
2,4-di	8.2×10^{-11}	6.8×10^{-12}
2,4,6-tri	5.2×10^{-11}	4.0×10^{-12}
2,3,7,8-tetra	3.0×10^{-11}	2.1×10^{-12}
1,4,6,8-tetra	3.7×10^{-11}	2.7×10^{-12}
2,3,4,7,8-penta	1.9×10^{-11}	1.3×10^{-12}
1,3,4,7,8-penta	2.2×10^{-11}	1.5×10^{-12}
Octa	4.3×10^{-12}	2.5×10^{-13}

[a]Above solid compound
[b]From the adsorbed state

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3. SYNTHESIS OF PCDFs

The possible routes for the synthesis of PCDFs in the laboratory have been reviewed by Garå *et al.*[3] Hereby, eight possible routes are described. Most of the synthetic routes mentioned result in mixtures of PCDF isomers. Of these eight synthetic routes, only the palladium (II) acetate-promoted cyclization of diphenyl-ethers, usually yields one isomer.

Recently 24 PCDF-isomers were synthesized by using ortho-hydroxy polychlorinated biphenyls (PCBs) as precursors. The ortho-hydroxy PCBs were synthesized by two routes by use of (1) a diazo coupling of chlorinated anisidines and symmetrical chlorinated benzenes and (2) diazo coupling of chlorinated anisoles. PCDF-isomers could be obtained with high purity by means of these syntheses (Figure 2).[4]

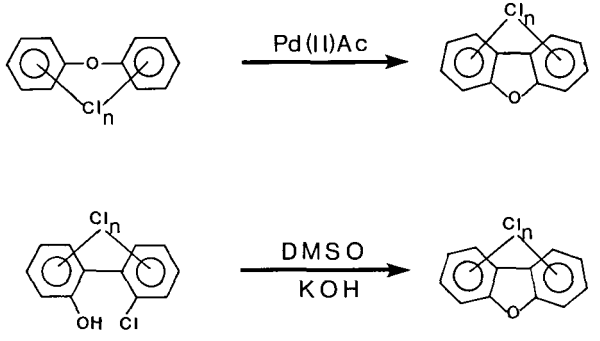

FIGURE 2 Two synthetic routes for PCDF formation with high isomeric specificity.[3,4]

The pyrolysis of PCBs and polychlorinated diphenyl ethers (PCDPEs) yields PCDF mixtures via several competitive mechanisms. Although isomeric mixtures arise by this synthetic route, this method has been frequently used to prepare qualitative PCDF standards because it is fast and safe. The different mechanisms hereby involved are reviewed in Section 4.

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4. FORMATION BY PYROLYSIS

4.1 Polychlorinated Biphenyls (PCBs)

Pyrolysis of individual PCBs and commercial PCB mixtures by 550–650°C resulted in the formation of PCDFs. In this temperature range 0.2–1.6% of the PCBs were converted to PCDFs. An optimal PCDF formation was found at 550°C, while at 700°C PCDFs were completely destructed. By burning eighteen individual PCB isomers, Buser *et al.* elucidated four reaction pathways for PCB conversion to PCDFs (see Figure 3).

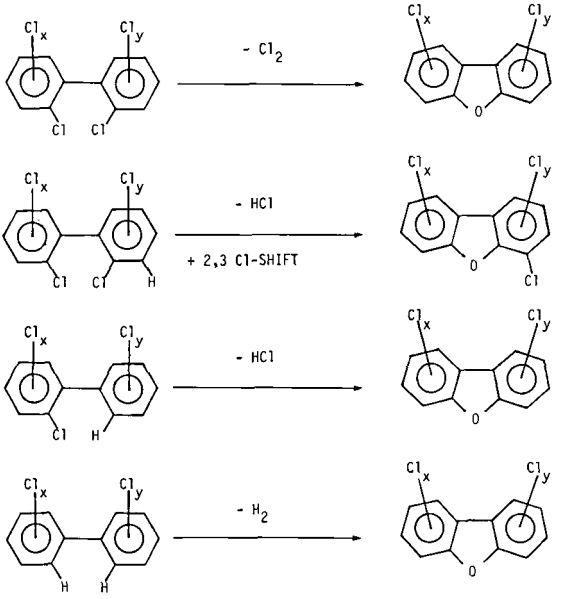

FIGURE 3 Mechanisms of PCDF-formation by pyrolysis of PCBs.[7]

The four mechanisms included:[5,6,7]

i) an ortho Cl_2-loss
ii) an ortho HCl-loss
iii) an HCl-loss with a Cl-shift from the 2 to 3 position
iv) an ortho H_2-loss

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By means of these four reaction mechanisms, it was possible to synthesize all 38 TCDF-isomers from their corresponding PCB congeners. These syntheses produced amounts of PCDFs which were sufficient for qualitative PCDF standards.[8] Pyrolysis of Aroclor 1254 and 1260 yielded a large number of TCDF and PnCDF isomers, whereby the toxic 2,3,7,8-TCDF, 2,3,4,7,8- and 1,2,3,7,8-PnCDF were present in considerable quantities. In fact, the 2,3,7,8-TCDF was found to be a major tetra-isomer.[6]

The facile conversion of PCBs to PCDFs could lead to environmental risks from transformer and capacitor fires, if PCBs are present as a heat transfer medium. Elevated PCDF levels have been found in soot samples which were collected after transformer fires in Canada, the U.S.A. and Sweden.[9,10,11,120,121] After an accidental fire in a Swedish transformer station, a PCDF level in the exploded capacitor was measured which was approximately 75 times the original PCDF concentration. In this accident the direct environment of the transformer building was also analysed, but elevated PCDF levels were only found in and on the exploded capacitor.[9] Moreover, in the U.S.A. a complete building was contaminated with PCDF-containing soot from an overheated transformer.[10,12] Detailed analysis of the soot from this building by Rappe and Buser revealed a total level of 2160 ppm PCDFs versus 20 ppm PCDDs.[12] The transformer fluid consisted of PCBs and polychlorinated benzenes, and both types of compounds are known to form PCDFs under pyrolic conditions.[5,6,7,13] The highly toxic 1,2,3,7,8- and 2,3,4,7,8-PnCDF represented 14.4 and 2.2% of the total PCDF content in the soot, respectively, whereas the 2,3,7,8-TCDF isomer represented about 0.5% of the total PCDF level.

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4.2 Polychlorinated Diphenyl Ethers (PCDPEs)

Under the same pyrolytic conditions described for PCBs, three reaction mechanisms were found for the conversion of PCDPEs to PCDFs. PCDF formation occurred primarily via the loss of ortho-H_2 and ortho-HCl. After the pyrolysis of one octa-chlorinated PCDPE sample, the loss of ortho-Cl_2 was found (see Figure 4).

In the temperature range from 500 to 700°C, the highest PCDF yield was found by 600°C. The PCDF yield also depended on the number and position of chlorine-atoms and varied from 1.7 to 4.5%.[14]

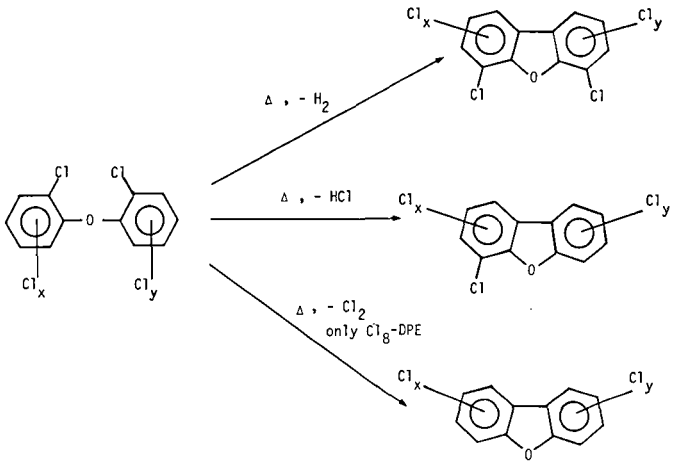

FIGURE 4 Mechanisms of PCDF-isomers by pyrolysis of PCDPEs.[14]

4.3 Polychlorinated phenols (PCPs)

In a combustion experiment in which chlorophenates were dissolved in water and then sprayed on birch leaves or wood shavings, no PCDF formation was found. The original PCP mixtures contained 40–50 PCDF isomers, and this number was reduced to ten after the burning. Some of the isomers found after the pyrolysis, however, were not present in the original chlorophenates. Based on these experiments and negative results of pure PCP-micro-pyrolysis, it was concluded that the PCDF-formation was a result of impurities in the commercial mixtures.[15] As commercial PCP formulations often contain PCDPEs, these impurities could have been the source of PCDF formation (see Figure 4).

4.4 Polychlorinated benzenes

PCDF formation was observed after micropyrolysis of tri-, tetra- and penta-chlorobenzenes at 620°C in the presence of air.[13] Furthermore, the general mechanism for PCDF and PCDD formation from chlorinated benzenes is given in Figure 5. The number of chlorine atoms in the formed PCDF was, in general, equal to 2m, 2m-1 or

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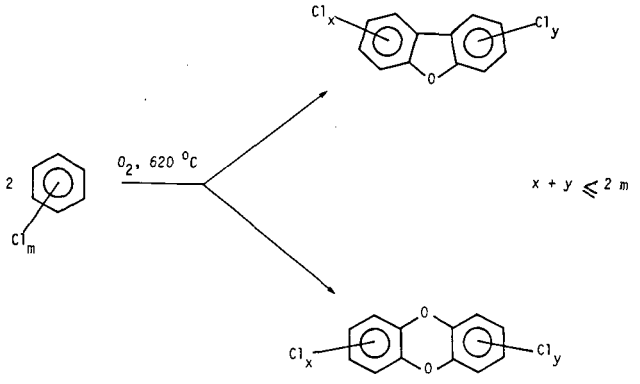

FIGURE 5 General mechanism for PCDF and PCDD formation from chlorinated benzenes.[13]

2m-2, whereby m is the number of chlorine atoms of the used chlorobenzenes. A combined chlorobenzene mixture of 7 tri-, tetra- and penta-isomers was pyrolized and resulted in a total of 58 PCDF-isomers in the tetra- to octa-range. The structure of 33 of these isomers could be identified. The highly toxic 2,3,7,8-TCDF, 2,3,4,7,8- and 1,2,3,7,8-PnCDF were not present as major components.

It is suggested that PCDF formation occurs via PCP intermediates which react further with chlorobenzenes to form PCDPEs before PCDF formation. This assumption could only partly be supported, because only PCPs were present in the pyrolysates, and PCDPEs could not be detected.

4.5 Phenoxy Herbicides

A 2,4,5-T formulation was combusted at different temperatures. The original concentration of PCDF was highest for TCDF (0.13 μg/g) and lower for the higher chlorinated isomers. An incomplete combustion of this formulation at 675°C produced the highest PCDF concentrations with 8.0, 7.1 and 2.8 μg/g, TCDF, PnCDF and HxCDF, respectively. No correlation could be found during these experiments between PCDF formation and temperature or combustion transit time.[16]

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4.6 Metals as Catalysts

Information about the catalytic function of metals in PCDF formation is scarce. When Aroclor 1248 was heated in the presence of oxygen for one week, the addition of $FeCl_3$ and Cu-Fe powder increased the formation of DiCDF and TrCDF at 270°C.[17] Individual PCB-isomers and PCB mixtures which were heated in the temperature range from 200–350°C for seven weeks yielded higher amounts of PCDFs when Fe was present.[18]

5. FORMATION AND MECHANISM IN MUNICIPAL AND INDUSTRIAL INCINERATORS

5.1 Occurrence of PCDDs and PCDFs in Fly Ash and Flue Gas

PCDFs have been found on fly ash as well as in flue gas from municipal and industrial incinerators in Europe, Canada and the U.S.A.[5,19,20,21] Concentrations of PCDF-homologues in flue gas have been found to range from approximately several hundred ng/m^3 in The Netherlands up to as high as 2.6 μg/m^3 in the U.S.A.[22,23] Both measurements have been listed in Table III.

TABLE III
PCDF concentrations found in flue gas from municipal refuse incinerators in The Netherlands and the U.S.A. in μg/m^3.[21,22]

	Netherlands	U.S.A.
TCDF	0.140	2.6
PnCDF	0.236	1.6
HxCDF	0.432	1.8
HpCDF	0.272	2.2
OCDF	0.035	0.17

Based on the flue gas measurements of both municipal incinerators, no correlation could be found regarding the distribution of the different homologues. However concentrations of PCDF in flue gas appeared to be considerably higher than those found for

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PCDDs.[21,22,23,24] Furthermore, measurements of PCDDs and PCDFs in Italy did not show any particular ratio between both groups of compounds.

Although HpCDD and OCDD were often major PCDD classes in individual samples, this trend could not be found for PCDFs. The average stack of PCDDs and PCDFs was almost equal for both classes.[24] Moreover, toxic 2,3,7,8-TCDF, 1,2,3,7,8- and 2,3,4,7,8-PnCDF isomers were first reported as major constituents in European fly ash, in contrast with their PCDD-congeners.[5] Recent analysis by Rappe[120] and the authors indicated however, that 2,3,7,8-TCDF was only a minor component. In Table IV the number of isomers detected in flue gas are compared with the number of isomers found for each group. The total number of PCDF isomers found in flue gas and fly ash is considerably higher than PCDDs, namely 45 PCDF isomers versus 29 PCDD isomers.

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TABLE IV
Detected PCDF and PCDD isomers in flue gas from municipal refuse incineration.[21]

	Number of detected isomers/Number of possible isomers	
	PCDD	PCDF
Tetra	11/22	17/38
Penta	8/14	12/28
Hexa	8/10	11/16
Hepta	1/2	4/4
Octa	1/1	1/1

However, it should be considered that the total possible number of PCDF isomers is higher than for PCDDs, because of the asymmetrical structure of the PCDFs. Nonetheless, the ratio of number of isomers detected/number of possible isomers is almost equal for PCDDs and PCDFs, that is 0.39 and 0.33, respectively.[21]

The contribution of each PCDF-isomer group to the total in fly ash was followed for four weeks in a Dutch municipal incinerator. The contribution of each group appeared to be rather stable during this period. These data are listed in Table V.[25]

TABLE V
Variation of PCDF content of fly ash in one municipal incinerator in The Netherlands.[25]

a. Content of fly ash (in ppb) Isomer group	Sample				b. Contribution of distinct group of isomers to total Sample					
	1	2	3	4	1	2	3	4	mean	*s*
TCDD	81	56	54	41	7.1%	5.0%	5.6%	3.2%	5.2%	1.6%
P5CDD	252	173	182	159	22.1	15.4	19.0	12.6	17.3	4.1
HCDD	406	388	326	381	35.6	34.5	34.1	30.1	33.6	2.4
H7CDD	321	385	288	458	28.1	34.2	30.1	36.2	32.2	3.7
OCDD	82	124	106	226	7.2	11.3	11.1	17.9	11.9	4.4
Total	1142	1126	956	1265						
TCDF	147	50	111	68	15.5%	7.1%	12.9%	7.3%	10.7%	4.2%
P5CDF	239	153	196	180	25.1	21.6	22.7	19.4	22.2	2.4
HCDF	345	282	361	357	36.6	39.8	41.8	38.5	39.1	2.3
H7CDF	202	202	177	291	21.1	28.5	20.5	31.4	25.4	5.4
OCDF	18	21	18	31	1.9	3.0	2.1	3.3	2.6	0.7
Total	951	708	863	927						

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5.2 Formation and mechanisms

Lustenhouwer *et al.* and Choudhry *et al.* have extensively reviewed the formation of PCDDs and PCDFs in incineration processes.[25,26,124] It is suggested that PCDF and PCDD formation during these combustions is partly a result of *de novo* synthesis of these compounds, and that the formation of these compounds is probably not mainly the result of the conversion of PCBs, PCDPEs, PCPs and chlorinated benzenes already present in the waste before the combustion.

The hypothesis states that PCPs and chlorinated benzenes, which are formed during the combustion, are important intermediates for PCDF and PCDD formation. The possible routes of formation of these compounds are shown in Figure 6.

According to this hypothesis there are three pathways for PCDF formation:[26]

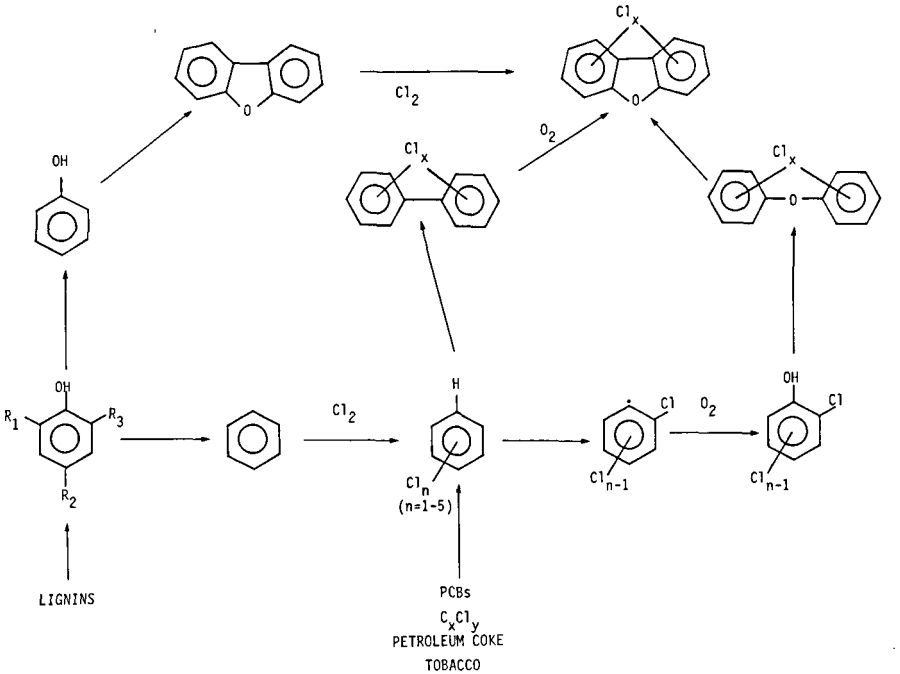

FIGURE 6 Speculative routes of PCDF-formation in municipal and industrial incinerators.[26]

i) via phenol with a possible chlorination after the dibenzofuran formation,

ii) via chlorinated benzenes with PCBs as intermediates,

iii) via chlorinated benzenes and PCPs with PCDPEs as intermediates.

Although PCBs are regularly mentioned as precursors of PCDFs[25,26,27] there seems to be very little evidence that PCBs are indeed key intermediates. The very small amounts of PCBs found in several combustion experiments makes this very unlikely.[21] However, chlorinated benzenes and PCPs can be found in high concentrations in flue gas and fly ash of incinerations. Therefore, both compounds or one of them might be precursors or reaction-intermediates during PCDF formation, although a direct correlation

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between these compounds and PCDF formation has not been found.[21]

Pilot scale incinerations indicated that waste burning of lignin-like structures in the presence of chlorine donors (e.g. PVC and HCl) yielded considerable amounts of PCDFs and PCDDs.[22] Combustion of vegetables yielded PCDFs and PCDDs in the μg/g range, with PnCDF, HxCDF and OCDF concentrations found to be higher than their corresponding dioxin congeners. The possible source of the PCDDs and PCDFs was attributed to a potential breakdown of phenolic structures of the vegetables, whereby these formed phenols were subsequently chlorinated and resulted in PCDF and PCDD formation.[28] Furthermore, combustion of pinewood at 600°C with hydrochloric acid as Cl-donor yielded 3015 ng PCDF per gram wood burned. PCDD formation in this experiment, however, was considerably lower. The highest PCDF concentrations were found for the hepta-isomers.[21] Consequently, these experiments indicate that waste burning of vegetables and wood can result in PCDF formation if a suitable chlorine-donor is present.

In Italy a distinct increase in PCDF and PCDD release was noted when the combustion temperature of the incinerator was relatively low, i.e. 500°C. This low temperature was probably caused by a high moisture content of the waste, possibly effected by the presence of many vegetables and humid weather.[24] The total emission increased during an 8 hour period to 2 grams for each class. This indicates that the composition of the waste as well as external factors, e.g. atmospheric conditions, could influence PCDD and PCDF emissions from municipal incineration. Moreover, industrial incineration of PCBs, PCPs, PCDPEs and chlorinated benzenes could also result in considerable amounts of PCDFs, if the incinerating temperatures are too low (see paragraphs 4.1, 4.2, 4.3, 4.4).

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6. FORMATION AND DEGRADATION BY PHOTOLYSIS

6.1 Formation from PCDPEs

The formation of PCDFs from lower chlorinated PCDPEs by photolysis has been examined in detail.[29,30] Irradiation experiments with wavelengths between 290–310 nm performed on a number of

mono-, di-, tri- and tetra-isomers of PCDEs in methanol, resulted in the formation of numerous PCDFs with lower chlorinated PCDPEs being major products.

No difference in photoproducts was found in hydrocarbon and methanol solutions.[30] When acetone was added as a photosensitizer to the substrate with PCDPEs, the PCDF yield increased greatly. Irradiation in pure acetone resulted in a yield varying from 46–76% for the lower chlorinated PCDFs.[31] When ortho-Cl was present in the PCDPE molecule, no reductive dechlorination occurred in acetone, but formation of PCDFs prevailed. If two PCDF-isomers were formed from the PCDPE, they were both formed by irradiation in acetone, e.g. 2′,3,4-PCDPE yielding 1,2- and 2,3-DiCDF.[31]

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6.2 Formation from PCBs

Irradiation experiments were performed with a F40B1 lamp indoors and in summerlight outdoors during 168–200 hours with eight PCBs. From 2,5-Cl_2-PCB and 2,2′,5,5′-Cl_4-PCB, 2-MCDF was found as a photoproduct with an approximate 0.2% yield.[32] In another experiment, five orthochlorinated PCBs were irradiated during 7 days resulting again in 2-MCDF from 2,5-Cl_2 and 2,2′,5,5′-Cl_4-PCB.[33] After photolysis of 2,2′,5,5′-Cl_4-PCB in a methanolic solution for 88 hours, trace amounts of DiCDF were detected.[30] Other irradiation experiments with PCBs did not yield PCDF[34,35] and it has been suggested that this was a result of rapid decomposition under energetic (254 nm) conditions or because of the presence of photo sensitizers.[36]

6.3 Formation from Polychlorinated Benzenes

Irradiation experiments (290–310 nm) performed with polychlorinated benzenes in mixtures of aqueous acetonitrile in the presence of phenols resulted in PCDF formation, with a $\leqq 1\%$ yield. The formation of PCDFs occurred through the formation of ortho-substituted PCDPEs as intermediate photoproducts. Experiments were performed by photolysis of three tetra-, penta- and hexa-chlorobenzenes with phenol and one tetra-chlorobenzene with 3,4-dichlorophenol. Based on the identified intermediate photoproducts, several mechanisms were postulated (Figure 7).[37]

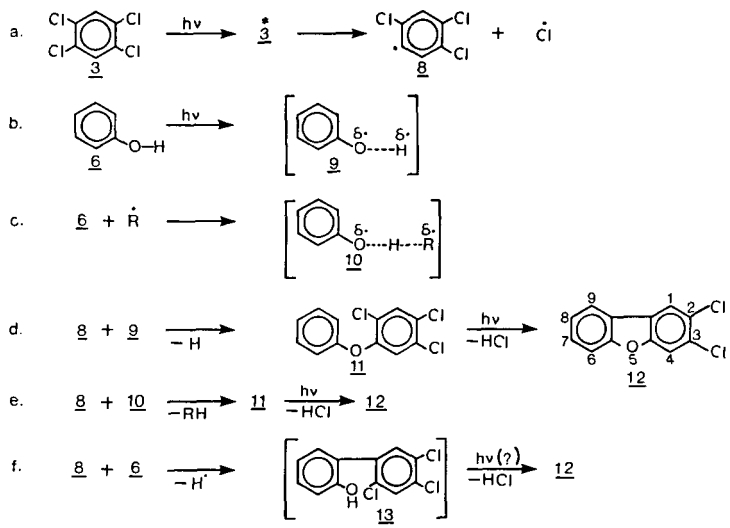

FIGURE 7 Mechanism of photochemical PCDF-formation from chlorinated benzenes and phenol in aqueous acetonitrile.[37] (Reprinted from *Chemosphere.*)

6.4 Photochemical Degradation of PCDFs

UV- and γ-irradiation of OCDF yielded considerable amounts of lower chlorinated PCDFs. After 4 hours of UV-radiation, HpCDF and HxCDF were the major photoproducts, while 10% of OCDF remained unchanged. After 24 hours only 2% OCDF was unchanged and HxCDFs appeared to be major photoproducts. γ-Irradiation also yielded HpCDF and HxCDF as major products. A difference was noted between HpCDF-isomers formed in both irradiation experiments, but isomeric structures were not determined.[38]

The photochemical degradation was faster in methanol than in *n*-hexane, and the decomposition rate of OCDF was similar to that of 2,8-DiCDF. However, a difference was found between UV-irradiation of 2,8-DiCDF in methanol and on a thin film. The former experiment produced MCDF and DiCDF, while the latter yielded TrCDF besides MCDF. Acetone did not function as a photosensitizer during the photodegradation of 2,8-DiCDF, but 4,4′-dichlorobenzophenone did. Photolysis of OCDF in methanol, a benzene/

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hexane mixture and on a thin film produced lower chlorinated PCDFs in all experiments.[36]

By studying the UV-photolytic products of eight TCDFs of known structure, a set of guidelines could be compiled for the prediction of photoproducts of TCDF.[39] These were formulated by Mazer and Hileman as follows:

1) Chlorines on the same aromatic ring tend to stabilize the loss of a chlorine from that ring.

2) Vicinal chlorines stabilize the loss of a particular chlorine (i.e. the greater the number of adjacent chlorines about a given chlorine, the greater the likelihood of initially losing that particular chlorine).

3) Given an equal number of vicinal chlorines, the 3-chlorine will be lost before the 2-chlorine.

By using photolytic dechlorination of known PnCDFs, TCDF structures could be confirmed which were pyrolytic products of PCBs. By using this technique and cross-correlation pyrolysis, all 38 TCDF-isomers could be identified and confirmed.[8]

7. OCCURRENCE IN COMMERCIAL PRODUCTS

Except for laboratory purposes, PCDFs are not commercially used. They are formed as unwanted by-products during the production of PCBs, PCPs and herbicides.[40,41,42,43,44,45,46] Occurrence of PCDFs in commercial polychlorinated naphthalenes should be more thoroughly investigated as reports concerning this are contradictory.[47]

7.1 PCBs

Commercial PCB-mixtures from European, North-American and Japanese origin have been found to contain PCDFs at ppm levels.[41,47] The European PCBs, Phenoclor DP 4–6 and Clophen A 30–50 had higher PCDF concentrations than the Aroclor 1242–1260 series produced in the U.S.A. The Japanese Kanechlor 300 to 600 products also contained PCDFs at ppm levels. In Tables VI and VII the PCDF concentrations in different commercial mixtures are listed. Major PCDF isomers found in PCB mixtures were 2,3,6,8-, 2,3,7,8-

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TABLE VI
Concentrations of PCDFs in commercial PCB mixtures in μg/g.

PCB	TCDF	PnCDF	HxCDF	Country
Aroclor 1248 (1969)[a]	0.5	1.2	0.3	USA
Aroclor 1254 (1969)[a]	0.1	0.2	1.4	USA
Aroclor 1254 (1970)[a]	0.2	0.4	0.9	USA
Aroclor 1254[b]	0.25	0.70	0.81	USA
Aroclor 1254 (Lot KK602)[b]	0.05	0.10	0.02	USA
Aroclor 1260 (1960)[a]	0.1	0.4	0.5	USA
Aroclor 1260 (Lot AK3)[a]	0.2	0.3	0.3	USA
Aroclor 1260[b]	0.3	1.0	1.10	USA
Aroclor 1016[a]	n.d.[c]	n.d.	n.d.	USA
Clophen A 60[a]	1.4	5.0	2.2	German Federal Republic
Clophen T 64[b]	0.3	1.73	2.45	German Federal Republic
Phenoclor DP-6[a]	0.7	10.0	2.9	France
Prodolec 3010[b]	1.08	0.35	0.07	France
Mitsubishi (used)[b]	4.00	3.30	0.53	Japan

[a]Data from Ref. 48.
[b,c]Data from Refs. 47

TABLE VII
Concentrations of PCDFs in Kanechlor in μg/g.[55]

Kanechlor	TCDF	PnCDF	HxCDF	HpCDF	conc. μg/g
300	+	+			1–1.5
400	+	+			17–18
500		+	+	+	2.5–4
600	+	+	+	+	3–5

and 2,3,6,7- TCDF, 1,2,4,7,8-, 1,2,3,7,8- and 2,3,4,7,8-PnCDF, 1,2,3,4,7,8-, 1,2,4,6,7,8- and 1,2,4,6,8,9-HxCDF, 1,2,3,4,6,7,8- and 1,2,3,4,6,8,9-HpCDF.[47] The highly toxic 2,3,7,8-TCDF and 2,3,4,7,8-PnCDF were found to be PCB constituents in concentrations varying from 0.08 to 0.83 μg/g PCB. In Aroclor 1242 the 2,3,7,8- and 2,3,4,7,8-isomers presented a large percentage of the total PCDF content, 43 and 19% respectively.[48] (See Table VIII). In the modern replacement of Aroclor 1242, i.e. Aroclor 1016, these PCDF isomers were not found.[49]

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TABLE VIII
Concentrations of 2,3,7,8-TCDF and 2,3,4,7,8-PnCDF in μg/g PCB in four commercial mixtures.[48]

	Aroclor 1248	Aroclor 1254	Kanechlor 200	Kanechlor 500
2,3,7,8-TCDF	0.33	0.11	0.10	0.19
2,3,4,7,8-PnCDF	0.83	0.12	0.10	0.08

7.2 PCPs

Nineteen PCP samples (technical, analytical grade and the Na-salt) were analyzed for their PCDF content. PCDF concentrations decreased in the sequence OCDF ~ HpCDF > HxCDF > PnCDF ~ TCDF. The technical formulations contained much higher concentrations than those of analytical quality. The PCDF contents found in these formulations are summarized in Table IX.[44] The lower chlorinated phenols also contained PCDFs in the μg/g range. This was found for 2,4,6-tri- and 2,3,4,6-tetrachlorophenol formulations (Table X).[50, 51]

Three commercial PCP mictures, 2,4,6-tri-, 2,3,4,6-tetra- and pentachlorophenol were analyzed for their isomeric PCDF content. The same major TCDF, PnCDF, HxCDF and HpCDF isomers were found in all samples. These isomers were 1,2,4,6,8-PnCDF, 1,2,3,4,6,8-, 1,2,4,6,7,8- and 1,2,4,6,8,9-HxCDF, 1,2,3,4,6,7,8- and 1,2,3,4,6,8,9-HpCDF.

The complete list of isomers found in these three PCP mixtures is shown in Table XI. The 2,3,7,8- and 2,3,4,7,8-isomers were not found as major isomers in the mixtures.[50, 52] It should be mentioned, however, that not all PCDF isomers were available when this study was done.

Because PCPs are widely used as insecticides, fungicides and antimicrobic agents, the PCDFs in PCPs could be transferred to the human food chain. It was found that out of 15 gelatine samples, 10 contained HpCDF and OCDF. As rawhides are some times treated with PCPs for preservation, the PCDF content of the gelatine might be caused by the use of PCP-treated hides.[53] The higher chlorinated HxCDF, HpCDF and OCDF were also found to be present in latex nipples. This PCDF content might be a result of pentachlorophenol treatment on the plantations were latex is collected.[54]

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TABLE IX
PCDF content in technical and analytical pentachlorophenol formulations.[44]

	TCDF	PnCDF	HxCDF	HpCDF	OCDF
1) Analytical quality (ppm)	<0.02–0.45	<0.03–0.13	<0.03–4.1	<0.1–13	<0.1–8.6
2) Technical quality (ppm)	<0.02–0.20	0.03–0.65	9.1–39	44–320	24–300

1) 8 samples
2) 11 samples

TABLE X
PCDF concentrations in $\mu g/g$ in PCPs.[50]

	PCDFs					
	Tetra-	Penta-	Hexa-	Hepta-	Octa-	∑PCDFs
2,4,6-Trichlorophenol, Sweden	1.5	17.5	36	4.8	—	60
2,4,6-Trichlorophenol, U.S.A.	1.4	2.3	0.7	<0.02	—	4.6
2,3,4,6-Tetrachlorophenol, Finland	0.5	10	70	70	10	160
Pentachlorophenol, U.S.A.	0.9	4	32	120	130	280
Pentachlorophenol, U.S.A.	—	—	30	80	80	190
Pentachlorophenol, U.S.A.	<0.4	40	90	400	260	790
Pentachlorophenol, Germany	—	—	0.03	0.8	1.3	2.1

TABLE XI
Isomers detected in three commercial polychlorinated chlorophenate samples.[52]

	2,4,6-Tri	2,3,4,6-Tetra	Penta
2,4,6,8-TCDF	+	–	–
2,3,6,8-TCDF	+	–	–
2,3,7,8-TCDF	+	–	–
1,2,4,6,8-PnCDF	+	+	+
1,3,4,7,8-PnCDF	+	–	–
1,2,3,7,8-PnCDF	+	–	–
2,3,4,6,8-PnCDF	+	–	–
2,3,4,7,8-PnCDF	+	–	–
1,2,3,4,6,8-HxCDF	+	+	+
1,2,4,6,7,8-HxCDF	+	+	+
1,2,4,6,8,9-HxCDF	+	+	+
2,3,4,6,7,8-HxCDF	+	–	–
1,2,3,4,6,7,8-HpCDF	+	+	+
1,2,3,4,6,8,9-HpCDF	+	+	+

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7.3 Herbicides

The presence of TCDF and PnCDF was found in Herbicide Orange, which is a mixture of the *n*-butyl ester of 2,4-D and the *n*-butyl ester of 2,4,5-T (approx. 50%/50% w/w).[45,46] From five recently produced 2,4,5-T esters and three Herbicide Orange formulations, PCDFs were only found in the latter. One TrCDF, four TCDF and one PnCDF were detected at a total level of 0.7 μg/g. However, none of the TCDF-isomers was the 2,3,7,8-isomer. Two older 2,4,5-T ester formulations, from 1967, contained 0.11 and 0.15 μg TCDF/g, both not the 2,3,7,8-isomer.

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8. OCCURRENCE IN BIOTA

Because PCDFs are released in the environment, these compounds could be expected in the food chain as by e.g. PCBs. Although analysis on PCDFs in living organisms is scarce, there are currently several positive findings of PCDFs which indicate its presence in wildlife.

Fish samples from midwestern and northern USA contained PCDFs with 4 to 7 chlorine atoms. Those fish samples containing PCDFs were contaminated with PCBs up to concentrations as high as 2 ppm.[56]

The first isomer-specific analysis on PCDFs was done by Rappe and co-workers. They analyzed the fat of the snapping turtle and grey seal.[57] The 2,3,7,8 congeners were present as major isomers. The fat of the snapping turtle had the highest concentration, i.e. a total of 3 μg PCDF/g fat. The 2,3,4,7,8-PnCDF concentration in the snapping turtle was approximately 75% (620 pg/g) of the total PnCDFs, while 2,3,7,8 TCDF was the only TCDF present at 45 pg/g. Based on the isomeric structures found in both samples, it was concluded that the PCDF contamination was a result of direct PCB contamination.

The PCDF content in fish samples collected in the Laurentian Great Lakes was found to be as high as 290 ppt. TCDF, PnCDF and HxCDF presented the majority of PCDFs in tissue, with TCDF content being as high as 40% of the total PCDF content. Again the 2,3,7,8 congeners were the major isomers, especially 2,3,7,8-TCDF and 2,3,4,7,8-PnCDF.

Low levels of PCDFs in fish were found from an isolated lake, remote from direct pollution sources. This might indicate atmospheric transport with rain-out of PCDFs parallel to PCB distribution. A comparison of PCDF content from sediment and fish in one of the lakes revealed a distinct difference in isomeric composition. PCDDs and PCDFs were also found in biological samples from the Baltic Sea recently.[122]

In sediment HpCDF represented most of the PCDFs, while in fish tissue this was the case for the lower chlorinated isomers.[58]

A similar difference in compositions between sediment and fish tissue was found for samples from the Hudson and Housatonic Rivers. HpCDF and OCDF were the dominating congeners present in sediment, but in fish tissue the TCDFs and PnCDFs were major congeners.[59]

Information on PCDF pollution from Europe is even more limited than for North America. In The Netherlands, mice from a heavily polluted area contained up to several ppb PCDFs in their liver with major isomers being 2,3,4,7,8-PnCDF and 2,3,7,8 TCDF. Analysis of livers of cormorants, a fish-eating bird, from one of the larger inland lakes in The Netherlands revealed the presence of 2,3,7,8-TCDF and 2,3,4,7,8-PnCDF, with up to several hundreds ppts found for the penta-isomer.[60]

Recently Rappe analyzed six human, breast milk samples from Germany and Sweden. All these samples contained measurable amounts of PCDFs and PCDDs. 2,3,4,7,8-PnCDF was present at a 1 ppt level.[61]

9. PHARMACOKINETICS

9.1 Uptake and retention

Mammals Birnbaum[62,63] and Decad[64,65] examined the uptake, accumulation and clearance of ^{14}C-labelled 2,3,7,8-TCDF in rats, rhesus monkeys, guinea pigs and two strains of mice after IV and oral application. After a single IV dose of 0.1 μmol/kg (30.6 μg/kg) for the rats, monkeys and mice and 0.02 μmol/kg for the guinea pigs, the tissues which retained most of the 2,3,7,8-TCDF were the liver, adrenals, adipose tissue and skin.

Rats also received an oral dose of 1 μmol/kg, and the distribution

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of 2,3,7,8-TCDF in the body was similar to the IV dose.[62] Considerable differences were found by the disposition of 2,3,7,8-TCDF in two strains of mice. In both C57BL/6J and DBA/2J mice the liver was the primary storage organ, but the TCDF concentration found in the C57BL/6J strain was 1.5 times higher. Although the administered 2,3,7,8-TCDF concentrations were equal for both strains, the percentage of the administered dose found in adipose tissue was much higher for DBA mice than for the C57 strain. The difference between both strains can be explained by the 72% higher adipose tissue content in DBA mice compared to the C57 strain. Because of this higher adipose tissue content DBA mice accumulate 2,3,7,8-TCDF more readily than C57 mice.[65] In the guinea pig, there was no significant difference in body distribution between IV and oral single dose administration. The liver, fat and adrenals were the major disposition sites.

A second experiment with guinea pigs involved a weekly oral dose of 1 μg 2,3,7,8-TCDF/kg administered for 6–7 weeks. The total uptake of 2,3,7,8-TCDF was very high (5.6–5.6 μg/kg). A difference was found in body distribution in both experiments. This might be explained by a higher fat mobilization by guinea pigs in the single dose experiment, resulting in the detection of a higher liver and lower fat percentage of the total dose.[64]

In Table XII the percentages of the total oral and IV doses found in liver and fat are given. At least 90–95% of the measured radioactivity in the liver and fat was the parent compound in all four species.

After the "Yusho" oil intoxication in Japan the isomer-specific accumulation in rats, monkeys and mice has been the subject of several investigations.[66, 67, 68, 69]

A mixture of TCDF-, PnCDF- and HxCDF-isomers with unknown isomeric composition was administered to ICR mice.[67] After administration of 0.5 mg PCDFs the highest concentrations were found in fat, liver and spleen. Because of the lack of isomeric information of the mixture, the retention potency of the individual isomers could not be determined.

Kuroki[66] administered 10 mg PCDFs/kg intraperitoneally to rats and monkeys. The mixture consisted of 1,2,7,8- and 2,3,7,8-TCDF, 1,2,4,7,8-, 2,3,4,7,8- and 1,2,3,7,8-PnCDF and one HxCDF. After 5 days isomer-specific retention decreased in the sequence HxCDF, 2.3.4.7.8-

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TABLE XII
Retention of 2,3,7,8-TCDF in liver and fat of four mammalian species

Species	% of total dose in liver		% of total dose in fat		Days after administration	Dose in μmol/kg	Reference
	Oral	Intravenous	Oral	Intravenous			
Guinea pig	17.50	29.30	46.07	56.90	3	0.02	64
Rat	5.02	7.0	4.64	13.8	3	0.1	62
Monkey	—	1.7	—	6.1	21	0.1	63
C57BL/6J Mouse	—	22.7	—	2.9	3	0.1	65
DBA/2J Mouse	—	16.8	—	22.3	3	0.1	65

PnCDF, 1,2,3,7,8-PnCDF, 2,3,7,8-TDCF. The other two isomers were not detected in the liver. The same mixture was given daily for 26–32 days to Rhesus monkeys at concentrations of 1.25 and 2.5 μg/kg. As by rats a specific retention occurred in the liver. The sequence of retention in the liver of the monkeys was 2,3,4,7,8-PnCDF > 1,2,4,7,8-PnCDF > 1,2,3,7,8-PnCDF ~ HxCDF > 2,3,7,8-TCDF. The difference in retention of the isomers between the rat and the monkey in these experiments might be explained by the different modes of administration, possibly a result of a less effective uptake after an oral dose compared to an intraperitoneal dose. Moreover, fly ash extracts containing large numbers of PCDDs and PCDFs were mixed with food and orally administered to rats. By comparing PCDFs and PCDDs, it was found that the retention of PCDFs in the liver was higher. The retention of the PCDF isomer groups increased with number of chlorine atoms from 4 to 6. Only one PnCDF, the 2,3,4,7,8-isomer, had a high retention. Of the HxCDF isomers which were retained, three accounted for about 85% of the total found. One of these three was probably the 2,3,4,6,7,8-HxCDF.[70]

In another experiment 13 individual isomers were intraperitoneally administered (1 or 10 mg/kg) to rats. The selective retention resembled the previous experiments and the most accumulated PCDFs were 2,3,4,7,8-, 1,2,3,7,8-PnCDF and 1,2,3,4,7,8-HxCDF. Compared to these isomers 2,3,7,8-TCDF was less accumulated. From the 2,3,4,7,8-PnCDF dose 65–70% was retained in the liver after 5 days, versus 3.8% for 2,3,7,8-TCDF. If 2,3,4,7,8-PnCDF is compared to 2,3,7,8-TCDF, the retention of the former isomer was approximately 20 times higher.[69]

Female ddN-mice were fed 18 days after mating or 14 days after delivery with a mixture of 0.6 ppm PCDFs containing 3 TCDF-, 3 PnCDF- and 1 HxCDF isomer. Of the total intake, only 0.003% of the PCDFs were transferred to the fetuses.

Furthermore the PCDF transport via the mother milk was much higher than the transport to the fetuses. After 2 weeks, 0.14% of the total uptake by the mother was found in the sucklings. Although transport across the placenta occurs, the transport via the mother milk was much more important. In the experiment and as expected, the livers of the mother mice had high concentrations, i.e. approximately 5.2% of the total PCDF-dose. 2,3,7,8-TCDF was the only

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tetra-isomer which had a high retention. Two PnCDFs were found with high liver retention, one of them being identified as 2,3,4,7,8-PnCDF.[68]

For 160 days cows received food mixed with pentachlorophenol contaminated with higher chlorinated dibenzo-*p*-dioxins and dibenzofurans.[71] From the six chlorinated dibenzofurans only two isomers were retained in the liver. One of the isomers was tentatively identified as 1,2,3,4,6,7,8-HpCDF and the other was OCDF. There was a clear ratio difference between both compounds. In pentachlorophenol the hepta-isomer was 1/3 of the OCDF concentration, but in the livers the hepta-isomer was 2/3 of the OCDF concentration. Based on this experiment it seems that HpCDF accumulates better than OCDF, probably due to the poorer absorption of OCDF from the intestinal tract. With increasing number of chlorine atoms, the lipophilic character of polychlorinated dibenzo-*p*-dioxins and dibenzofurans also increases. Higher chlorinated PCDDs and PCDFs have very low solubilities in polar solvents, making it difficult to be absorbed by the intestinal tract.[72]

Birds Only one experiment has been carried out with avian species. Mallards were fed 5–100 mg Aroclor 1254/kg for approximately one year.[73] Aroclor 1254 is a PCB mixture which contains 0.11 μg 2,3,7,8-TCDF/g PCB and 0.12 μg 2,3,4,7,8-PnCDF/g PCB.[48] At the end of this period, only 3% of the ingested PCDFs was still present in the Mallards. However, this 3% was based on the whole lipid content in the body which makes it impossible to specify body distribution in general.[73]

Fish Juvenile Atlantic salmons received 2.5, 5.7, 2.8 and 9.1 μg/g 2,8-DCDF, TrCDF, TCDF and OCDF, respectively.[74] Only OCDF was later found in the fish, but the fate of the lower chlorinated PCDFs was not discovered.

Humans In 1968 more than 1500 persons were poisoned in Japan by rice oil contaminated with a PCB mixture which also contained PCDFs. This poisoning is commonly called "Yusho disease". In 1979 a similar poisoning occurred on Taiwan.[75] Especially the Yusho-patients have been the subject of thorough research, and it is now

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established that the PCDFs in the patients have caused most of the toxic symptoms.

An average Yusho patient ingested approximately 20 mg PCDF mixture, from which 0.25 mg was 2,3,7,8-TCDF.[76,77] The PCDF mixture consisted of a large number of TCDF-, PnCDF- and HxCDF-isomers. PCDFs which were retained in the liver were 2,3,7,8-, 2,3,6,8-TCDF, 2,3,4,7,8-, 1,2,4,7,8-, 1,2,3,7,8-PnCDF, 1,2,3,4,7,8- and 1,2,3,6,7,8-HxCDF. Moreover, a high retention was found for 2,3,4,7,8-PnCDF and 1,2,3,4,7,8-HxCDF in the livers of "Yusho" patients. A higher concentration of 2,3,6,8-TCDF than 2,3,7,8-TCDF was found in the liver, while in the original Yusho-oil 2,3,7,8-TCDF had a higher concentration.[78]

The liver of one Yusho-patient, 44 months after ingestion of the rice oil, had the following percentages of the ingested amounts: 2,3,6,8-TCDF 0.37%, 1,2,4,7,8-PnCDF 0.006–0.03% and 2,3,4,7,8-PnCDF 0.9%. The 2,3,7,8-TCDF isomer was not found in the liver after this period. In the liver of another Yusho-patient, 2,3,4,7,8-PnCDF and 1,2,4,7,8-PnCDF were still found nine years after exposure.[79]

One of the patients who died two years after the poisoning in Taiwan was examined for PCDF content. Liver and intestinal fat were the major disposition sites in the body. Two major toxic isomers, as in the Yusho poisoning, were found to be 2,3,4,7,8-PnCDF and 1,2,3,4,7,8-HxCDF. In the liver 2,3,7,8- and 2,3,6,8-TCDF were detected, but both isomers had a minor retention compared to 2,3,4,7,8-PnCDF and 1,2,3,4,7,8-HxCDF.[80]

When PCB and PCDF concentrations of 5 Yusho patients were compared with healthy persons 1 to 9 years after the poisoning, PCB concentrations were only a few times higher whereas PCDF concentrations were higher by 2 to 3 orders of magnitude in Yusho patients. This indicates that at least some PCDF-isomers could have a higher retention in humans than some PCBs.[81,82] Humans which were occupationally exposed to polychlorinated phenols had maximum concentrations as high as 400 pg PCDFs/g in their blood. The major isomer in the blood was 1,2,3,4,6,7,8-HpCDF, which is also a major PCDF in tetrachlorophenol.

9.2 Excretion and Metabolism

Mammals When rats were fed Aroclor 1254 which normally con-

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tains 2,3,7,8-TCDF and 2,3,4,7,8-PnCDF, first indications showed that 2,3,7,8-TCDF could be excreted by the urine.[84]

Birnbaum and Decad examined the half-life of 2,3,7,8-^{14}C-TCDF in rats, rhesus monkeys, mice and guinea pigs.[62,63,64,65] These data are summarized in Table XIII.

Administration of 0.02 μmol 2,3,7,8-^{14}C-TCDF to guinea pigs resulted in 6.6% excretion of the total dose in urine and feces in 9 days. In the feces 90% of the excretion was 2,3,7,8-TCDF, while in the urine more than 90% consisted of one or more polar metabolites. No 2,3,7,8-TCDF was detected in the bile, which might indicate that a passive excretion occurs via the feces.[64] The calculated half-life in guinea pigs in this experiment was approximately 20 days. A single dose of 0.1 μmol 2,3,7,8-^{14}C-TCDF which was given orally or intravenously to Fisher 334 rats, was excreted very rapidly from the body, with a whole body half-life of less than two days. The major route of excretion was via the feces, which contained 85% of the radioactivity of the administered dose. Excretion via the urine was less important and contained after 7 days no more than 6% of the total dose. The rapid excretion of 2,3,7,8-TCDF in the rat compared to guinea pigs, is a result of a more efficient metabolism, because more than 99% of the excreted 2,3,7,8-TCDF in the rat were metabolites.[62]

In the rhesus monkey a whole body half-life of 8 days was found after an intravenous dose of 0.1 μmol/kg 2,3,7,8-^{14}C-TCDF.[63] Twenty-one days after dosage 43% of the total dose of 2,3,7,8-TCDF was excreted in the feces versus 8% in the urine. In the feces and urine two metabolites of unknown structure were present, but in the feces also traces of the parent compound were found. The majority of the excreted radioactivity from the feces, bile and urine were metabolites. The capacity of metabolizing and excreting 2,3,7,8-TCDF in rhesus monkeys appears to be intermediate between this capacity in guinea pigs and rats. After an intravenous dose of 0.1 μmol 2,3,7,8-^{14}C-TCDF/kg in two strains of mice (see 9.1), the whole body half-life depended largely on the amount of adipose tissue in the strain. The estimated half-life for DBA/2J mice in fat was 4–5 times higher than for C57BL/6J mice. The half-life in the liver was for both strains equal. The major excretion route for both strains was via the feces. A minor part of the excreted radioactivity in urine and feces was parent compound, while metabolites were predominant in both excretion routes.[65]

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From the above experiments it appears that rat, mice and rhesus monkeys can metabolize 2,3,7,8-TCDF more easily than 2,3,7,8-TCDD. For these species the major way of excretion is via the feces in the form of metabolites. Because the guinea pig lacks the more efficient metabolism of the former species, it has a whole body 2,3,7,8-TCDF half-life comparable with that of 2,3,7,8-TCDD. When a 0.5 mg PCDF mixture containing two TCDF-, four PnCDF- and four HxCDF-isomers was intraperitoneally administered to mice, the estimated half-life for this mixture was approximately 2 weeks. Because the isomeric structures of this mixture were not known, these results were difficult to interpret. However, one PnCDF and two HxCDFs from this mixture were slowly or not measurably excreted.[67]

Humans Humans, who were intoxicated by "Yusho" oil, consumed a mixture containing more than 40 PCDF-isomers. In the livers of these patients the number of measurable isomers decreased to approximately 10. Although all the isomers which had a high retention in the livers of these patients were identified, the structures of only a limited number of excreted isomers could be identified. These isomers were 2,3,6,7-TCDF, 2,3,4,6,7-, 1,2,6,7,8-, 1,2,3,4,8-PnCDF and 1,2,3,4,6,7-HxCDF. These isomers all have two vincinal hydrogen-atoms in common.[78] Similar results were also found for a patient who ingested toxic rice-bran oil on Taiwan in 1979.[80]

Metabolism in Mammals The remarkably fast excretion of 2,3,7,8-TCDF in rats, mice and rhesus monkeys as compared to 2,3,7,8-TCDD (see Table XIII) must be the result of a more facile,

TABLE XIII
Half life (in days) of 2,3,7,8-TCDD and 2,3,7,8-TCDF in four mammalian species.[62, 63, 64, 65]

Species	2,3,7,8-TCDD[a]	(dose μg/kg)	2,3,7,8-TCDF	(dose μg/kg)
Guinea pig	30	(2.0, IP)	20	(6, IV)
Rat	31	(1.0, oral)	<2	(30.6, IV)
C57BL/6J Mouse	17	(100, IP)	2	(30.6, IV)
DBA/2J Mouse	37	(100, IP)	4	(30.6, IV)
Rhesus monkey	—	(—)	8	(30.6, IV)

[a]Data from Ref. 85

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metabolic conversion of 2,3,7,8-TCDF. It was suggested that a stereochemical factor causes this difference. Compared to 2,3,7,8-TCDD, the 2,3,7,8-TCDF molecule has a more strained carbon-oxygen bridge, which could facilitate enzymatic attack.[72]

An attempt has been made to elucidate metabolism of the chlorinated dibenzo-furans.[86] In the rat, 2-MCDF, 2,8-DiCDF, 2,3,8-TrCDF were easily metabolized, and several monomethoxy-, dimethoxy- and methylthioderivatives were found in urine and/or feces. As the polar extracts were methylated to analyse the metabolites with GC/MS, it is not known whether the above mentioned compounds were truly methylated metabolites or *in vivo* monohydroxy, dihydroxyglutathione or mercapturic acid conjugates.

Because five monomethoxy metabolites were found from 2,8-DiCDF, metabolic conversion includes a chlorine NIH-Shift. No metabolites were found from OCDF, although the parent compound was determined in the liver after administration. Besides the metabolites already mentioned, polar compounds were found which resulted from breaking the carbon-oxygen bridge.[87] Compared to the PCDDs the dibenzofurans showed a greater variety of metabolites.[88] The metabolism of 2,3,7,8-TCDF was studied and four major metabolites were found, namely one trichloromethoxy-, two trichlorodimethoxy- and one tetrachloromethoxy-dibenzofuran. The methoxylated compounds probably originated from hydroxylated forms via methylation. One minor metabolite was tentatively identified as a tetrachlorodimethoxy-PCB probably formed by the ether-bridge cleavage. Based on this experiment it was concluded that ether cleavage did not present a major metabolic pathway.[89]

By means of the Hückel Molecular Orbital Calculation for PCDDs and PCDFs the possible position of the hydroxy groups in both types of molecules was predicted. Based on these calculations it was suggested that preferential enzymatic attack on a dibenzofuran molecule would be on the 1,2 position, whereas in the dibenzo-*p*-dioxin molecule the 2,3 position would be favoured. These calculations included 1,2,3,4- and 2,3,7,8-TCDF.[90]

Administration of PCDFs can also influence the biological half-life of PCBs in liver and adipose tissue. PCDFs and PCBs were separated from the contaminated "Yusho" oil and were again dissolved in pure rice oil which was administered to mice. The presence of PCDFs had little influence on the total biological half-

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life of PCBs, but the half-life of three individual PCBs was markedly reduced.[91] These results suggested that PCDFs can apparently activate metabolism of certain PCB isomers. This might be a result of the enzyme-inducing effect which is similar for PCDFs and certain PCB isomers (e.g. 2,3,3′,4,4′-Cl_5-PCB and 3,3′,4,4′-Cl_4-PCB). See Section 10.1.

10. BIOCHEMICAL INDICATIONS

10.1 Enzyme Induction

The polychlorinated dibenzofurans show a 3-methylcholanthrene type of enzyme-induction, the most obvious result being an increase of cytochrome P-448 and arylhydroxycarbonhydroxylase (AHH).[92] In this respect the PCDFs show similarity with the PCDDs.

In the rat the ED_{50} of 2,3,7,8-TCDF for AHH was determined by a 0.5 μg/kg/day dose administered for three days. This value was approximately one-fifth of that for 2,3,7,8-TCDD.[92] The ED_{50} value for 2,3,7,8-TCDF in rat hepatoma cell cultures was determined on 13 pmol/plate while the value for 2,3,7,8-TCDD was 0.14 pmol/plate. Other isomers tested in this experiment which showed AHH inducing capacities were 2,3,8-TrCDF, 1,3,4,7,8-PnCDF, 1,2,4,7,8-PnCDF and 1,2,3,4,6,8,9-HpCDF. These isomers showed inducing effects ranging approximately 20–340 lower than that found for 2,3,7,8-TCDF.[93]

In chickens, both 2,3,7,8-TCDD and -TCDF induced AHH, but in contrast with 2,3,7,8-TCDD the TCDF congener did not induce glucuronyl-transferase.[94] Induction of Ala-synthetase by 2,3,7,8-TCDF was not found in this experiment, however, induction of this enzyme was found in a chicken-embryo experiment at approximately 5×10^{-11} mol/egg.[95]

Thirteen PCDF isomers were tested in rats for their enzyme induction capacities. 2,3,7,8-TCDF, 2,3,4,7,8- and 2,3,4,6,7-PnCDF had the highest inducing effect, approximately 10 times the AHH level detected in the control group at an intraperitoneal dose of 1 mg/kg. Some isomers which produced a strong AHH induction also increased the cytochrome P-448 content in rat livers twofold.

Benzphetamine-demethylase was not induced by any of the tested isomers, but was strongly inhibited by 2,3,7,8-TCDF, 2,3,4,7,8- and

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1,2,3,7,8-PnCDF and 1,2,3,4,7,8-HxCDF. However the former two isomers were dosed at 1 mg/kg, while the latter two were dosed at 10 mg/kg, thereby making it difficult to compare the inhibiting effect of these four isomers. The cytosolic enzyme DT-diaphorase was induced by the same isomers which had strong AHH-inducing capacities. A dose-response experiment showed that 2,3,7,8-TCDF as well as 2,3,4,7,8-PnCDF still had a significant inducing effect on AHH and DT-diaphorase at 1 μg/kg.[69] In Table XIV the known AHH-inducing PCDFs are listed with their estimated AHH-inducing capacities relative to 2,3,7,8-TCDF. The estimated values have been derived from experiments in which AHH induction of PCDFs was listed and could be compared with that for 2,3,7,8-TCDF in the same experiment.

TABLE XIV
The estimated AHH activity for a number of active TCDF isomers.[69, 93, 96]

TCDF isomers	AHH-inducing capacity
2,3,7,8	1
2,3,4,6,7	1
2,3,4,7,8	0.75–1
1,2,3,4,7,8	0.75
2,3,6,7	0.5
1,2,6,7,8	0.5
1,2,3,4,6,7	0.5
1,2,7,8	0.4
1,2,3,7,8	0.2–0.6
1,3,4,7,8	0.05
2,3,8	0.005
1,2,3,4,6,8,9	0.004
1,2,4,7,8	0.003

AHH-enzyme assays with PCDFs showed that substitution with chlorine atoms on three of the four lateral positions of the PCDF molecule was necessary.[69, 93, 96] As with PCDDs, AHH-induction by PCDF is strongest when all four lateral positions are occupied.

Furthermore, 2,3,7,8-TCDF possesses a AHH-inducing capacity which is of the same order of magnitude as that of 2,3,7,8-TCDD.[94, 96] In PCDFs substitution of the 2,3,6 and 7 positions produces an induction which is in the same order of magnitude as

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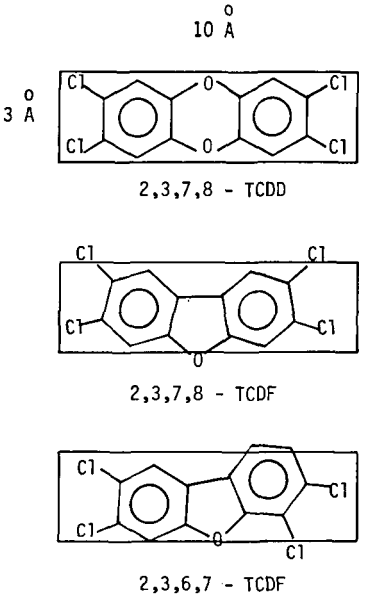

FIGURE 8 Structure resemblance for 2,3,7,8-TCDD, 2,3,7,8-TCDF and 2,3,6,7-TCDF.

found for 2,3,7,8-substitution. This was found for the pairs 2,3,6,7-/2,3,7,8-TCDF, 2,3,4,6,7-/2,3,4,7,8-PnCDF and 1,2,3,4,6,7-/1,2,3,4,7,8-HxCDF.[69] The explanation for the additional AHH-inducing pattern (2,3,6,7 in addition to 2,3,7,8) might be simple. By considering bond length and atomic radii in a PCDF molecule it becomes evident that 2,3,6,7-TCDF fits well or better into the (3×10 Å) TCDD receptor of Poland. See Figure 8.

From most of the evidence now collected, it appears that there is a clear correlation between toxicity of chlorinated arylhydrocarbons and their AHH-inducing capacities.[97] From this aspect it seems logical that PCDFs which have AHH-inducing capability can produce significant biologic or toxic effects.

Recently the inducing effect of a fly ash extract for ethoxyresorufin O-deethylase (EROD) was compared with AHH-induction. Comparable results were found for the induction of both enzymes which are found in rat hepatoma cells. By using these induction

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TABLE XV
Comparative bioassay analysis of fly ash extracts.[98]

Sample	∑ PCDDs (ng/g)	∑ PCDFs (ng/g)	Estimated dioxin content (ng/g) AHH (ng/g)	EROD (ng/g)	Receptor assay (ng/g)
1	918	2363	3.93	4.55	32
2	<0.5	<0.5	—	—	—
3	30.1	68.3	—	—	3.5
4	203.1	91.3	3.93	4.55	65.0
5	31.2	31.2	1.45	2.25	11

assays, the "active dioxin" content was estimated to be much lower than the actual PCDD and PCDF content of the fly ash extract. See Table XV.

10.2 Cytosol Receptor Binding

The toxicity of chlorinated arylhydrocarbons correlates well with their potential to induce AHH. It is believed, however, that the toxic effects of these compounds, including PCDFs, are not caused by AHH but that both AHH induction and the toxic symptoms are both secondary after an initial binding to a cytosolic receptor-binding protein.[97] At present only the relative protein-binding affinity of 2,3,7,8-TCDD, 2,3,7,8-TCDF, 1,2,3,7,8- and 2,3,4,7,8-PnCDF is known.[95] These are listed in Table XVI.

TABLE XVI
Cytosolic receptor binding affinity and AHH induction of PCDFs relative to 2,3,7,8-TCDD.[96]

Compound	Relative binding activity	Relative AHH induction
2,3,7,8-TCDD	100	100
2,3,7,8-TCDF	37	67
2,3,4,7,8-PnCDF	34	71
1,2,3,7,8-PnCDF	38	14
1,3,6,7-TCDF	Inactive	Inactive
2,4,8-TrCDF	Inactive	Inactive
2,4-DiCDF	Inactive	Inactive
2,8-DiCDF	Inactive	Inactive

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Although the cytosol-binding affinity of the three most toxic PCDF isomers has been established, this has not been determined for a large number of other TCDF, PnCDF and HxCDF isomers which cause AHH induction (cf. Table XIV). When a fly ash extract sample was tested for dioxin binding to the hepatic, cytosol receptor protein, it was found that binding was 45 times more active than could be expected based on the amount of 2,3,7,8-TCDD alone.[99] When the receptor assay was compared with the AHH and EROD assays, the calculated "active dioxin" content of fly ash was considerably higher in the receptor assay than in the others.[98] See Table XV.

Because fly ash of incinerators can contain a complex mixture of PCDDs and PCDFs, the question is raised regarding the contribution of PCDFs to the mixture's biochemical activity. From this aspect it seems noteworthy that two fly ash extracts were compared for the presence of some very biologically active PCDF and PCDD isomers. It was found that some highly active PCDF isomers were major PCDF constituents, while their PCDD analogues were found to be minor constituents in fly ash.[5]

Quantitative relationships were developed for a series of PCDDs and PCDFs regarding their structures and for their cytosol-binding and AHH-induction activity. By using regression analyses, calculations indicated that lateral substituents on a PCDD or PCDF molecule do not influence AHH-induction and receptor binding in the same way. The calculations also demonstrated that five structural factors influence the degree of AHH-induction and receptor binding. Cheney formulated the following factors as being:[100]

1) an orbital with the characteristics of the $4b_{1g}$MO in TCDD,

2) a bulky substituent in a longitudinal position,

3) a crowded nitro group occupying one of the lateral sites,

4) a dibenzofuran cyclic system,

5) an electronegative atom in a substituent attached to a lateral ring carbon.

Because PCDF showed unexpected, high AHH-induction and cytosol-binding activity, the particular influence of the dibenzofuran molecule on these activities as compared to the influence of the dibenzo-*p*-doxin molecule is especially interesting for this review.[101]

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11. ANIMAL TOXICITY AND CLINICAL SYMPTOMS

Rats

After a single intravenous dose of 30.6 μg 2,3,7,8-TCDF/kg or oral dose of 306 μg 2,3,7,8-TCDF/kg to male Fisher 344 rats, decreased weight gain, excessive hair loss and listlessness were observed after two days. However, after 3 weeks post-treatment the rats gained their normal weight and had no further, visible toxicological symptoms. In this experiment there was also no visible sign of atrophy of the thymus or spleen. The rapid recovery after intoxication was probably caused by the rapid 2,3,7,8-TCDF elimination in this strain of rats.[62]

In a CD rat strain a single oral dose as high as 1000 μg 2,3,7,8-TCDF/kg produced no intoxication symptoms and microscopic examination of the tissues revealed no abnormalities.[102]

Moreover, Sprague–Dawley rats received a diet containing 1 and 10 ppm PCDF for 4 weeks. The isomeric structures of the PCDFs were unknown, but the mixture contained two TCDF-isomers, four PnCDF-isomers and four HxCDF-isomers. Chloracne developed in the animals receiving the 10 ppm diet after one week. At both PCDF levels by the diet, a relative liver weight increase and a significant decrease of the absolute and relative weight of the thymus and spleen were observed. At both doses, severe atrophy of the thymus and increased serum cholesterol and liver lipid concentrations were also found.[103,104]

The same symptoms were developed by rats given orally 35 μg PCDFs/day (unknown structures) for 10 days. Additionally a vacuolation of liver cells, kidney glomerulus and adrenal cortex was observed.[105]

Wistar rats receiving a single 1 mg/kg IP dose of 2,3,7,8-TCDF, 2,3,4,6,7- and 2,3,4,7,8-PnCDF developed severe atrophy of the thymus. This effect was also produced by a single 10 mg/kg IP dose of 1,2,3,7,8-PnCDF and 1,2,3,4,7,8-HxCDF. These five isomers also had a strong enzyme-induction capacity. 2,3,6,7-TCDF and 1,2,3,4,6,7-PnCDF had only a slight effect on the thymus at 1 mg/kg, while 1,2,7,8-TCDF and 1,2,6,7,8-PnCDF had no effect at all. All the MC-type inducing isomers had an effect on the liver weight, except 1,2,7,8-TCDF. 2,3,7,8-TCDF and 2,3,4,7,8-PnCDF produced similar thymic atrophy and liver hypertrophy at a dose range from 1 μg/kg to 10 mg/kg.[69]

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Mice

Mice seem to be relatively resistent to PCDFs. A single oral dose of 6000 μg 2,3,7,8-TCDF/kg did not elicit any toxic symptoms in the C57BL strain. When the same dose was applied subcutaneously weight loss, hepatomegaly and thymus atrophy occurred which, however, did not result in death.[106]

Due to the strain-related body clearance of 2,3,7,8-TCDF, its toxicity is not expected to be equal for different strains of mice. As a result of the slower body clearance of 2,3,7,8-TCDF for DBA/2J compared to C57BL/6J mice, the toxicity of 2,3,7,8-TCDF can be lower for the latter strain.[65]

An oral dose of 30,100,300 μg 2,3,7,8-TCDF/kg was administered 22 times during thirty days and produced no clinical symptoms in the C57BL/6J strain. The administration of about 6 mg 2,3,7,8-TCDF/kg to this strain resulted in an effect on organ weights which was approximately 30–33 higher than for 2,3,7,8-TCDD.[106]

CF-1 mice receiving single oral, intraperitoneal or subcutaneous doses of a PCDF-mixture containing 42% TCDF, 54% PnCDF and 4% HxCDF developed no toxic symptoms during the first week. The lethal dose for this mixture (with unknown isomeric structures) was established as 300 mg/kg oral and 60 mg/kg intraperitoneal, and the time until death was 10–20 days. The lowest concentrations at which thymus/body weight, liver/body weight and body weight were influenced were 30 mg/kg for males and 67 mg/kg for females.[107]

Mice which were administered a mixture of 12% TCDF and 88% PnCDF at a dose of 10 or 100 μg/kg once a week during four weeks, showed only a thymus weight-effect at the highest dose for males.[108]

Guinea pigs

Guinea pigs which received a single dose of 1 μg 2,3,7,8-TCDF or 1 μg 2,3,4,7,8-PnCDF/kg showed a decreased weight gain. Like in other species, there was a time delay before death occurred. For a 10 μg/kg dose this time delay was approximately two weeks. When a higher dose, 15 μg 2,3,7,8-TCDF/kg or 30 μg 2,3,4,7,8-PnCDF/kg was administered, the time before death was shortened to 11.8 and 9.2 days, respectively. The symptoms caused by both isomers were similar as those found for 2,3,7,8-TCDD in guinea pigs. Some of the symptoms were reduction of thymus and body-fat.[106]

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Guinea pigs receiving a weekly dose of 1 μg 2,3,7,8-TCDF/kg during a period of six weeks suffered 30% mortality. In the same experiments animals receiving 1 and 0.5 μg/kg showed a significant reduction in thymus/body weight ratio. At both dosages no change of spleen/body weight ratio was found.[109]

A comparison was made between a single intravenous dose of 6 μg 2,3,7,8-TCDF/kg and approximately the same oral total dose equally distributed over six to seven weeks. The toxic symptoms in the six to seven week lasting experiment were much more severe than in the single IV dose experiment. The most important symptoms were loss of hair, listlessness and hydration. There were no signs of hepatomegaly or thymus atrophy.[64]

Rabbits

A mixture of TrCDF and TCDF isomers given as a single oral dose of 1 mg or 0.5 mg/kg produced liver necrosis. Application of this mixture to the rabbit's ear caused severe hyperkeratosis.[102,110]

Chickens

Chickens were fed daily 5 μg or 1 μg 2,3,7,8-TCDF/kg during 21 days. The 1 μg/kg dose did not affect the liver/body weight ratio, and only one out of six chickens died. At the higher dose 100% died, with an average time of death being 11.5 days. At both levels subcutaneous edema, ascites, hydropericardium and thymic atrophy occurred. At 1 μg/kg there was still a decrease in food consumption and body weight gain.[111]

The symptoms observed in the above experiment are equal to those of the "chicken edema" disease, which was caused by 1,2,3,7,8,9-HxCDD.[110,112] Because PCDFs can apparently produce the same toxic symptoms, this raises the question about what the contribution of PCDFs to the "chicken edema disease" was, since PCDF-isomers are often present in commercial pentachlorophenols.

Monkeys

The estimated LD_{50} for a single oral dose of 2,3,7,8-TCDF for rhesus monkeys was determined as 1000 μg/kg. A single oral dose of 500 μg/kg caused skin lesions and reduced weight gain. Between 500–1500 μg 2,3,7,8-TCDF/kg a dose related response was found for the

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toxic symptoms, which usually appeared after 7 to 10 days after administration of the single dose. The LD_{50} value of 2,3,7,8-TCDF was approximately 20 times higher than that found for 2,3,7,8-TCDD.[106]

In two chronic experiments, rhesus monkeys were fed 5 ppb and 50 ppb 2,3,7,8-TCDF-containing cakes for several months. Toxic symptoms from both doses appeared to be similar to those from a single dose of 2,3,7,8-TCDF or 2,3,7,8-TCDD. Death occurred at both levels, and it was calculated that the total uptake was 300 μg/kg (50 ppb) or 90 μg/kg (5 ppb). As the estimated LD_{50} value was considerably higher, it is assumed that chronically administered 2,3,7,8-TCDF at low doses is more damaging than a single high dose. During the 1 to 3 months feeding period, 2,3,7,8-TCDF appeared to be 10% as active as 2,3,7,8-TCDD at comparable doses. The most important toxic symptoms were weight loss, swelling of the eyelids, skin changes, hair loss, reduced physical activity, atrophy of the thymus, squamous metaplasia of the sebaceous glands, metaplasia of the stomach and bile duct mucosa and hypoplasia of the bone marrow. Although the toxicity of 2,3,7,8-TCDF is of the same magnitude as 2,3,7,8-TCDD, the recovery after 2,3,7,8-TCDF intoxication proceeded more rapidly than recovery after 2,3,7,8-TCDD intoxication.[113,114]

Administration of a single intravenous dose of 30.6 μg 2,3,7,8-TCDF/kg, a dose far below the LD_{50} value, still produced significant pathological changes in the rhesus monkey. Among these were changes already mentioned above.[63]

A comparative study was done with Kanechlor 400 with cynomolgus monkeys, which received a 20 week-long diet. PCDF-free KC 400, distilled KC 400 with 400 ppm PCDFs and PCDF-free, distilled KC 400 were administered. Monkeys which received the distilled KC 400 with PCDF showed decreased body weight, lower antibody production, liver hypertrophy and pathological changes. These symptoms were more pronounced than those caused by the other two groups.[115]

The oral LD_{50}-values for 2,3,7,8-TCDF, 2,3,4,7,8-PnCDF and 2,3,7,8-TCDD in the preceding species are listed in Table XVII.

Humans

The only reported exposure of humans to PCDFs regarded the Yusho and Taiwan food poisonings. As the rice oils were also

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TABLE XVII
Oral LD_{50} in μg/kg for 2,3,7,8-TCDF, 2,3,4,7,8-PnCDF and 2,3,7,8-TCDD in different species.

Species	2,3,7,8- TCDF	2,3,4,7,8-PnCDF	2,3,7,8-TCDD
Guinea pig	5–10[106]	3–10[106]	2[85]
Rat	1000[101]	?	22–45[116,a]
Mouse C57BL/6J	6000[106]	?	114–150[116]
Monkey	1000[106]	?	50[85]

[a]Spartan strain

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contaminated with PCBs, the symptoms found in these patients might not be caused by PCDFs alone.[47]

Considering the clinical symptoms of several species exposed to PCDF, which closely resemble those symptoms caused by PCDDs, it might be expected that symptoms found in humans after exposure to 2,3,7,8-TCDD will roughly resemble those from PCDFs. As by PCDDs the intensity of the symptoms will vary with the chlorine substitution pattern.

Some authors believe that PCDFs were mainly responsible for the Yusho and Taiwan poisonings. These statements are supported by the fact that workers occupationally exposed to PCBs did not show the same pathological changes as found in Yusho and Taiwan patients.[75] With this in mind, the strong enzyme-induction activity of 2,3,4,7,8-PnCDF and its extremely high retention in livers of Yusho patients may lend this isomer importance by the Taiwan and Yusho poisonings.

12. MUTAGENICITY, TERATOGENICITY AND CARCINOGENICITY

Information about mutagenicity and teratogenicity of PCDFs has been very limited. Three isomers, including 2,3,7,8-TCDF, were tested and results were negative. In mice teratogenic effects have been found for 2,3,7,8-TCDF at ppb levels. However, no information was available about carcinogenicity. Considering the chemical and

biological properties of PCDFs which can closely resemble those of PCDDs, thorough research should be carried out on PCDFs.

Mutagenic activity of 2,3,7,8-TCDF was tested on one strain of *Saccharomyces cerivisae* and on two strains of *Salmonella typhimurium*, whereby no activity was found in both species.[117,118] In the *S. typhimurium* strains, OCDF, 2,9- and 3,6-DiCDF were also non-mutagenic, and were also non-mutagenic when microsomal extracts with varying concentrations from both uninduced and induced rats were added to the experiments.[117]

Treatment of pregnant C75BL/6N mice with doses varying from 10 to 100 μg 2,3,7,8-TCDF/kg on gestation days 10–13 resulted in a dose-related increase in cleft palates and hydronephrotic kidneys in the fetuses.One hundred per cent teratogenicity occurred at levels which were not maternally toxic.[119]

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13. CONCLUSIONS AND SUMMARY

Polychlorinated dibenzofurans (PCDFs) is a group of compounds which resembles polychlorinated dibenzo-*p*-dioxins (PCDDs) in physical, chemical and biological behaviour. However, some distinct differences in formation and occurrence in the environment are found between both groups.

PCDFs can be formed from PCBs, PCDPEs and chlorobenzenes during combustion. Four mechanisms have been found for the formation of PCDF from PCBs. Because of the facile conversions of PCBs to PCDFs, the occurrence of fires in transformers containing PCBs can be a serious environmental hazard. Uncontrolled burning of PCBs, PCDPEs and chlorobenzenes, at too low temperatures, can also produce significant amounts of PCDFs.

PCDFs have been found in commercial PCB- and PCP-mixtures in concentrations up to ppm amounts. The highly toxic 2,3,7,8-TCDF, 1,2,3,7,8- and 2,3,4,7,8-PnCDF can be found in commercial PCBs, while in PCPs these isomers are minor or not measurable.

Fly ash and flue gas from municipal and industrial incinerators can contain a complex matrix of PCDFs. The most toxic isomers are usually found as minor components.

Photochemical formation of PCDFs can occur from PCBs, PCDPEs and chlorobenzenes. If a suitable photosensitizer, e.g.

acetone, is present, PCDF formed from ortho-substituted PCDPEs can have a very high yield.

The information about PCDF occurrence in living organisms is limited. However, PCDFs have been found in seal, fish, birds, adipose tissue and breast milk. Of special interest is the apparently high bioaccumulation potential of 2,3,4,7,8-PnCDF in several species. This isomer does have a very high retention in human livers and is now believed to be responsible for a number of toxic symptoms found in "Yusho" patients.

Most of the pharmacokinetical information is based on experiments with 2,3,7,8-TCDF. Body half-life for this isomer varied from 2 days for the rat to as high as 21 days for the guinea pig. Major storage of 2,3,7,8-TCDF occurred in the liver, skin and adipose tissue. With the exception of the guinea pig, most mammals more efficiently metabolize 2,3,7,8-TCDF than 2,3,7,8-TCDD. For metabolism the presence of two vincinal unsubstituted carbon atoms facilitates conversion to more polar metabolites. Furthermore cleavage of the oxygen-bridge and NIH-shift have been found as results of metabolic processes.

As by PCDDs, PCDFs show a 3-methylcholanthrene-type of enzyme induction. The enzyme-inducing capacity of 2,3,7,8-TCDF and 2,3,4,7,8-PnCDF is slightly lower than that found for 2,3,7,8-TCDD, but it is still in the same order of magnitude. Isomers having a 2,3,6,7-substitution pattern show an enzyme induction almost equal to those isomers with four lateral chlorine-substituents.

2,3,7,8-TCDF produces clinical symptoms in different species, which closely resemble those of 2,3,7,8-TCDD. However, these symptoms are largely species-dependent. The guinea pig is the most sensitive species, with LD_{50} values for 2,3,7,8-TCDF and 2,3,4,7,8-PnCDF in the same range as found for 2,3,7,8-TCDD.

Mice and rats are the least sensitive species, while the rhesus monkey shows an intermediate sensitivity. Rapid recovery after intoxication with 2,3,7,8-TCDF in the less sensitive species as compared to 2,3,7,8-TCDD, is probably caused by the more efficient metabolism and consequent excretion of 2,3,7,8-TCDF.

Information about mutagenicity and teratogenicity of PCDFs is very limited. Three isomers, including 2,3,7,8-TCDF, were tested and results were negative. In mice teratogenic effects have been found for 2,3,7,8-TCDF at ppb levels. No information was available for

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carcinogenicity. Considering the chemical and biological properties of PCDFs which can closely resemble those of PCDDs, thorough research should be carried out on PCDFs.

From an environmental point of view, the occurrence of 2,3,4,7,8-PnCDF should cause serious concern. The high biochemical activity and retention in mammals gives this isomer properties closely resembling those of 2,3,7,8-TCDD.

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