Fate of Octachlorodibenzo-p-dioxin in Small Ponds

by

Line Marcheterre

A thesis

presented to the University of Manitoba

in partial fulfillment of the

requirements for the degree of

Master of Science

in

Department of Soil Science (Chemistry of Pesticides)

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FATE OF OCTACHLORODIBENZO-P-DIOXIN IN SMALL PONDS

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ABSTRACT

On August 17 1983, two outdoor artificial ponds were treated with ^{14}C-O_8CDD at levels of 340 ng/L and 680 ng/L. The movement of the label was followed in air, water, fish, plants, and sediments for 652 days post-treatment; the ^{14}C-labelled material was analyzed qualitatively by HPLC and quantitatively by liquid scintillation counting. Volatilization was found to be a minor pathway for the loss of O_8CDD from the pond system. O_8CDD remained within the aquatic environment; it disappeared very rapidly from the water column and was taken up by fish and plants. The fish did not accumulate O_8CDD to the extent observed for the vegetation; both floating and rooted vegetation were unable to retain the label far longer. The sediment bed appeared to be the most important reservoir for O_8CDD; concentrations of O_8CDD in the sediment increased over time, up to 652 days post-treatment. The label seemed to be tightly complexed to the sediment as shown by the poor extraction efficiencies observed. The significance of these results may be that O_8CDD would not be appreciably available to fish or plants but would rather accumulate and persist in sediments of aquatic systems.

CONTENTS

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I INTRODUCTION

Dioxins have never been used by man as a commercial product and they are not formed naturally in the environment except via combustion processes in ultra trace quantities. They can occur in different concentrations in a number of useful chemicals such as pesticides. In this manner, they have been unintentionally introduced in the environment for many years (Korte, 1983).

Chlorinated phenols are manufactured in large amounts and have widely varying applications (Nilsson et al., 1978). Pentachlorophenol has been shown to contain important quantities of O_8CDD (Goldstein et al., 1977). When considering the environmental input of pentachlorophenol (in Canada, 2.5×10^6 kg a year), it becomes important to evaluate the fate of its impurities.

Cultivated terrestrial regions to where pesticides such as pentachlorophenol are regularly used are those in which the most profound effects are likely to be evident (Heckman, 1982). However, most persistent agricultural chemicals, along with their impurities, eventually find their way into nearby water bodies where they can accumulate and/or be transported throughout entire watersheds. In western Canada, O_8CDD has been detected in Tobin Lake sediments at approximately 50 parts per trillion (Webster et al., 1981). The Saskatchewan river system flows into Tobin Lake. O_8CDD has also been found in the Great Lakes (Stalling et

al., 1983; Czuczwa and Hites, 1984).

This research project was thus undertaken to investigate the distribution and persistence of O_8CDD in a realistic situation under field conditions. Outdoor artificial ponds were treated with ^{14}C-O_8CDD and samples of water, plants, fish and sediments taken at various times post-treatment and analyzed by HPLC and LSC. The objectives of this thesis were to determine the bioavailability of O_8CDD in a pond system, its degree of loss by volatilization and its fate in water, sediments, plants and fish.

II LITERATURE REVIEW

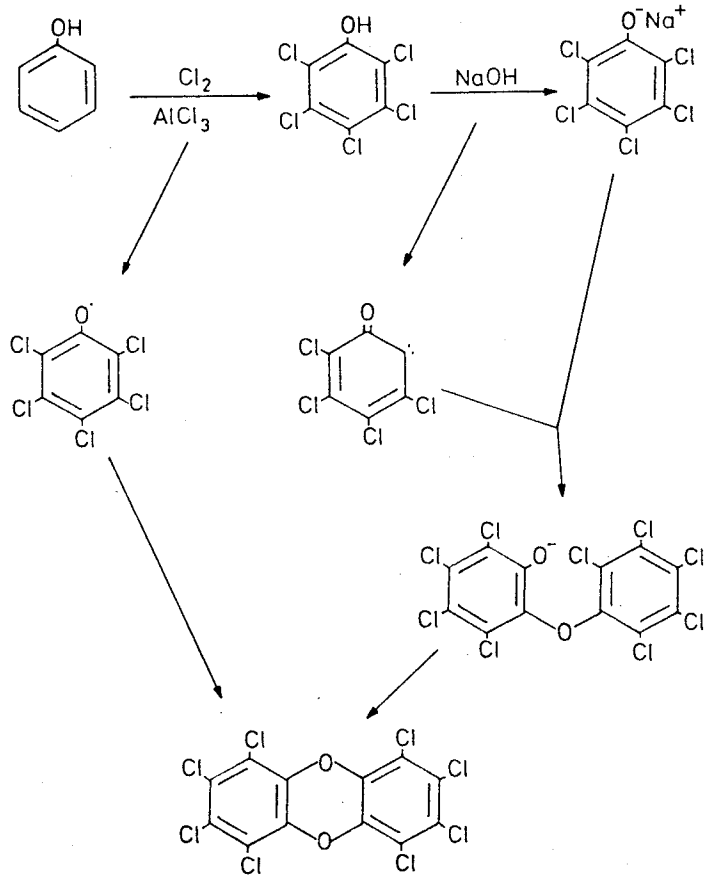
1.0 Formation and Occurrence of O_8CDD

1.1 Chemical Sources. The widespread occurrence of O_8CDD within the environment results from its presence as a contaminant of the wood preservative PCP (pentachlorophenol) as well as its formation in incinerators and other combustion processes. Photolytic formation of O_8CDD on wood treated with PCP has been reported but O_8CDD formation via this process was thought to be non important (Lamparski et al., 1980).

O_8CDD can be formed via the synthesis of PCP and the fungicide chloranil, 2,3,5,6-tetrachloro-1-4-benzoquinone (Esposito et al., 1980). PCP can be synthesized by the direct chlorination of phenol with an $AlCl_3$ catalyst (Scheme 1) or, as an alternative process, formed by the reaction of hexachlorobenzene (Scheme 2). The complex free-radical mechanism by which chloranil is formed results in the presence of short-lived intermediates, similar to those occurring as by product derivatives of PCP (Scheme 3). For these reasons, O_8CDD should be expected as a contaminant of chloranil.

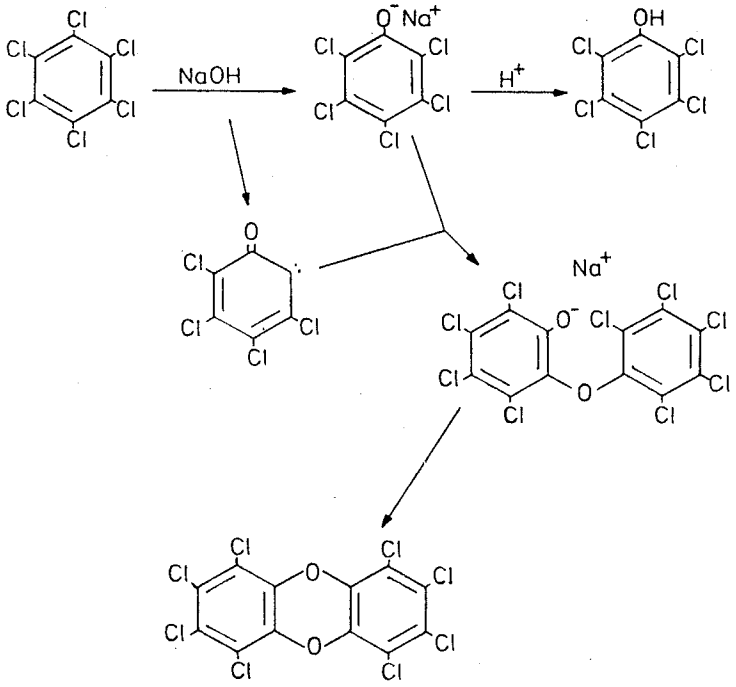
Scheme 1

O_8CDD Formation during Synthesis of Pentachlorophenol (PCP) from Phenol (Esposito et al., 1980).



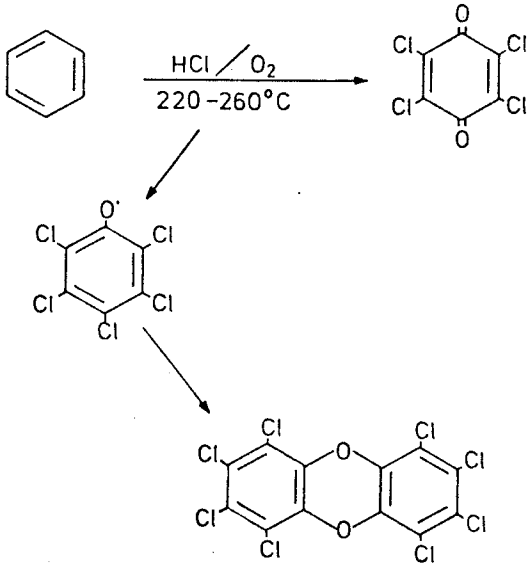
Scheme 2

O_8CDD Formation during Synthesis of PCP from Hexachlorobenzene (Esposito et al., 1980).



Scheme 3

O_8CDD Formation during Synthesis of Chloranil from Benzene (Esposito et al., 1980).



Chlorinated phenols and related materials, particularly PCP, could be the major source of total PCDD input to the Canadian environment (NRCC, 1981). In 1978, Canada's consumption of PCP was estimated to be approximately 2.5 X 10^6 kg a year. Its usage in Canada is mainly involved with wood treatment. In Western Canada, wood is treated by dipping in a tank of PCP, for short-term protection against mold and bacteria (Rao, 1978). Concentrations of O_8CDD in technical PCP on the Canadian market have ranged from 120 to 650 mg/kg, with an averaged concentration of approximately 370 mg/kg (Singh, 1981). Numerous analyses have confirmed the presence of O_8CDD in PCP (Esposito et al., 1980). Cull et al., (1984) recently published results of the analysis of 8 different samples of technical PCP obtained from 3 different analytical methods. Their results (Table 1) were in good agreement considering the difficulties involved in the clean-up and analysis. The typical concentrations of the higher chlorinated dioxin isomers in PCP are in the order $O_8CDD > H_7CDD > H_6CDD$ (Goldstein et al., 1977). A great number of analyses have confirmed that dioxins with less than 6 chlorine atoms are generally not detected in PCP (Esposito et al., 1980).

Table 1

PCP Contamination by O_8CDD (Cull et al., 1984)

Analytical method	Mean concentration of O_8CDD in technical PCP (µg/g)
GC-ECD	610 ± 13%
HPLC	585 ± 13%
GC-MS	472 ± 25%

1.2 Combustion Sources. With respect to the formation of PCDD's in municipal incinerators, Choudhry et al.(1982) have suggested that likely major routes of formation are from chemically related compounds such as chlorophenols, e.g., PCP, chlorobenzenes and PCB's. A number of studies in the USA, Europe, and Canada, have shown the presence of PCDD in flyash from incinerators (Dow Chemical, USA, 1978). Lustenhower et al.(1980) report PCDD concentrations in electrostatically precipitated flyash and flue gas particulate matter coming from 25 different installations in The Netherlands. O_8CDD concentrations ranged from 106 to 266 µg/kg in flyash samples and 310 µg/kg in particulate matter samples. In Canada, Clement et al., (1984) published work on the determination of PCDD's in incinerator effluents. They extracted and quantified as much as 50 µg/kg of O_8CDD in flyash from a number of Ontario sources.

1.3 Photolytic Formation. Lamparski et al.(1980) observed short term increases of O_8CDD concentrations in wood treated with purified PCP and exposed to sunlight. Dobbs and Grant (1981) found that it appears to be a question of time before seeing a decrease of O_8CDD concentrations in wood treated with PCP. They reported that the analysis of treated wood exposed outdoors for long periods showed no overall formation of O_8CDD. Cull and Dobbs (1984) reported that over a 2.5 year peri-

od, analysis of wood treated with technical PCP showed that photolytic dechlorination of O_8CDD to less chlorinated isomers was taking place. However, there was no discernible increase in O_8CDD concentrations, presumably because further photolytic reactions and volatilization competed effectively with the photolytic formation. Cull et al., (1983) reported that volatilization of O_8CDD from treated wood was a major pathway for its loss.

Considering the importance of Canada's consumption of PCP and knowing the great quantities of O_8CDD present in PCP as impurities, there should be some concern about an environmental buildup of O_8CDD. O_8CDD input to the environment can also result from its formation in combustion sources such as municipal incinerators which have been shown to be important mode of entries of O_8CDD to the environment. Field studies such as ours should bring some valuable information on the fate and dissipation of O_8CDD in an aquatic system, where persistent chemicals such as O_8CDD often accumulate.

2.0 Toxicity of O_8CDD

Toxicity data on O_8CDD is very scarce but not conflicting as it agrees on its weak toxic, teratogenic, and mutagenic effects. Schwetz et al., (1973) found that oral doses of 1 g/kg of O_8CDD did not cause death to female rats; oral doses of 4 g/kg to male mice were not

lethal either. Its acute toxicity was thus qualified as very low and LD_{50} estimated to be approximately 1000 mg/kg body weight. No external sign of toxicity (loss of body weight for example) were observed in any animals treated with O_8CDD. In a similar feeding experiment, 2,3,7,8-T_4CDD was lethal to male guinea pigs at 0.0006 mg/kg body weight and lethal to rabbits of both sexes at 0.115 mg/kg. O_8CDD was found to have a very low or nonexistent acnegenic activity in the rabbit. No signs of maternal toxicity were observed in pregnant female rats given 100 and 500 mg/kg-day of O_8CDD. However, O_8CDD caused embryotoxicity at 500 mg/kg-day as subcutaneous edema was observed among the fetal population. No such toxic effects were observed at 100 mg/kg-day. These authors concluded that the toxicity of O_8CDD is very low. Wassom et al., (1977/1978) hoped to answer many questions concerning the mutagenicity of 2,3,7,8-T_4CDD and other dioxin isomers as a result of their work. They reported that research on dioxins as potential mutagens was initiated because of their similarity to acridines, a class of known DNA intercalating agents. They tested O_8CDD on 5 different strains of Salmonella typhimurium and reported the octa isomer as a questionable mutagen in 2 strains and as a nonmutagen in 3 others. Goldstein et al.,(1977) studied the effect of PCP on the liver of female rats, (Table 2). The idea

Table 2

Contamination of Pure and Technical Grades of PCP with Chlorinated Dioxins (Goldstein et al., 1977).

		Pure	Technical
Phenols:	PCP	>99%	84.6%
	T_4CP	<0.1 ppm	3%
Dioxins:	T_4CDD	<0.1 ppm	<0.1 ppm
	P_5CDD	<0.1	<0.1
	H_6CDD	<0.1	8
	H_7CDD	<0.1	520
	O_8CDD	<0.1	1380

was to compare the effects of pure and technical grades of PCP on the development of hepatic porphyria and alterations in hepatic drug-metabolizing enzymes. All dietary levels of technical PCP; 20, 100, and 500 ppm, increased aryl hydrocarbon hydroxylase (AHH) activity (15 to 43 fold). Liver weight and liver/body weight ratios were also increased by technical, but not by pure PCP. The presence of dioxins in technical PCP was thought to be responsible for the toxic effects reported by Goldstein and coworkers. However, it is unlikely that O_8CDD was responsible for the hepatic effects of PCP because it is not lethal at doses 10^4 times the LD_{50} of 2,3,7,8-T_4CDD and because it does not induce AHH in the chick embryo (Poland et al., 1976). In 1979, Poland and coworkers demonstrated that there was an excellent correspondence between the relative toxicity of some PCDD isomers and their activity as inducers of AHH as well as

their binding to a cystolic receptor protein. On a scale of 0 to 100, O_8CDD was found to have a relative binding affinity and a relative biological potency of zero. In the same study, 2,3,7,8-T_4CDD was assigned a value of one hundred for both its relative binding affinity and relative biological potency. Goldstein et al., (1977) concluded that their results clearly show that technical PCP containing PCDD's produces a number of hepatic effects which cannot be attributed to PCP itself. In the light of the non-toxic effect of O_8CDD, NRCC(1981) has raised some concern about those studies involving oral ingestion of O_8CDD. As the report mentions it, its very low solubility might suggest that the vehicles used in oral dosing studies could result in its inefficient sorption and thus, the measured toxicity could be an artifact of the experimental procedure.

3.0 Pond studies

3.1 Ecosystem Studies with Dioxins. Persistent chemicals such as O_8CDD, once they enter the environment, tend to be transported to rivers and/or lakes where they can accumulate. To study the ecotoxicology of such chemicals, it is not advisable to contaminate natural bodies of water; furthermore, their value and complexity should limit their use for experimentation with persistent chemicals. It then becomes necessary to design aquatic ecosystems that will simulate natural conditions. These model ecosystems will represent a useful screening method for evaluating the

environmental toxicology, fate and distribution of chemicals under investigation. The design of model ecosystems can be quite varied and range in size from small aquaria and ponds to middle size quarries.

Very few ecosystem studies with dioxins have been reported; most of them involved 2,3,7,8-T_4CDD (Isensee and Jones, 1975; Ward and Matsumura, 1978; Yockim et al., 1978; Tsushimoto et al., 1982). Corbet et al. (1983) studied the fate of 1,3,6,8-T_4CDD in a model aquatic ecosystem, under field conditions.

Isensee and Jones (1975) designed small glass aquaria (25 x 5 x 17 cm) to simulate the usual mode of entry of 2,3,7,8-T_4CDD into water, and the simultaneous exposure of several organisms representing various constituents of natural food chains. They found that bioaccumulation took place in every level of the food chain and that the degree of accumulation was directly related to concentration of 2,3,7,8-T_4CDD in the water. They detected no metabolites.

In 1978, Ward and Matsumura investigated the persistence and metabolic degradation of 2,3,7,8-T_4CDD in lake sediment and water, under laboratory conditions. Most of the tetra isomer was found in the sediment from which it slowly disappeared ($t_{1/2}$= 600 days). They observed that metabolic activity was enhanced under conditions which stimulated microbial growth in the presence of sediments. Metabolites did not persist in sediments and were found to be released to the ambient water.

Yockim et al., (1978) used a recirculating static model ecosystem in attempting to more fully evaluate the environmental fate of 2,3,7,8-T_4CDD. Bioaccumulation ratios of 2-6 x 10^3 were obtained for various organisms (fleas, snails and algaes), results that were in agreement with those previously reported by Isensee and Jones (1975). These results indicated that the physical design of the ecosystem had little effect on the bioaccumulation ratios between their experiment and Isensee and Jones' study.

Tsushimoto et al., (1982) used outdoor enclosures of concrete construction to describe the movement of radiolabelled (^{14}C) 2,3,7,8-T_4CDD in a simulated aquatic ecosystem. As plant material died and settled down on the bottom sediments, they observed that the tetra isomer was persistent: 49.7% and 29.4% of the label remained after 12 and 25 months respectively. No metabolites were isolated or identified but were detected in plants and water, representing approximately 40% of the original ^{14}C.

Corbet et al. (1983) studied the fate of ^{14}C-1,3,6,8-T_4CDD in outdoor experimental ponds. Following its subsurface application, the label was lost through volatilization with highest amounts of volatilized ^{14}C-1,3,6,8-T_4CDD trapped during the first day. The remaining ^{14}C was found to accumulate in flora and fauna, as it was previously observed for 2,3,7,8-T_4CDD (Isensee and Jones, 1978; Yockim et al., 1978).

3.2 Studies with Other Persistent Chemicals. Many other persistent chemicals have been studied in model aquatic ecosystems. Among the number of studies that can be found in the literature, some chemicals have been chosen as reference compounds because their chemistry, toxicology and fate in the enviroment have been fully documented, or else, they are thought to be potentially hazardous and ask for immediate and thorough evaluation of their environmental fate is required.

Chlorinated hydrocarbons such as DDE and lindane were selected by Hamelink and Waybrant (1976) because they could bring valuable information on the fate and distribution of chlorinated hydrocarbons in a model aquatic ecosystem. They chose to use large flooded limestone quarries (91 x 41 x 15 m deep) instead of small farm ponds, pools, or aquaria primarily because the sediment-water and air-water interface values of the latter ecosystems often appear to be uncharacteristically high. Upon completion of a one year experiment, their results showed that the distribution of both chemicals in the system was controlled chiefly by sediment-water interactions. Bioconcentration factors for zooplankton, at equilibrium, averaged 33 for lindane and 4.5×10^4 for DDE; BCF values for bluegill were 768 and 1.1×10^5 respectively.

In light of the increasing importance of field studies in testing and validating mathematical models, Sugiura et al., (1984) studied the fate of 2,4,6-trichlorophenol, pentachlo-

rophenol (PCP), p-chlorobiphenyl, and hexachlorobenzene in outdoor experimental ponds. They demonstrated that the concentration changes of the test compounds in outdoor experimental ponds could be roughly estimated from a model, which described the movement and distribution of the compounds in the system, and the rate constants which had been determined in laboratory experiments.

Crossland and Wolff (1985) compared predictions of the fate and biological effects of PCP with observation in experimental outdoor ponds. PCP was chosen as a reference compound primarily because its chemistry, toxicology, and fate in the environment are well-documented. Evaporation, sorption, hydrolysis, biodegradation and indirect photo-transformation of PCP were not expected to be very important under conditions present in the pond system. The dominant loss process was predicted to be direct phototransformation. The calculated photodegradation rate constant ranged from 331 to 662 s^{-1}, values in good agreement with the observed rate which ranged from 216 to 489 s^{-1}.

4.0 Fate and distribution of O_8CDD in model aquatic systems.

In our study, small experimental outdoor ponds (5 x 4 x 0.5 m) were used to study the dissipation of O_8CDD from pond water and its subsequent distribution within fauna and flora present in the ponds.

Various removing, degradative and accumulating processes may be operative within the system of choice (Figure 1) (NRCC, 1981).

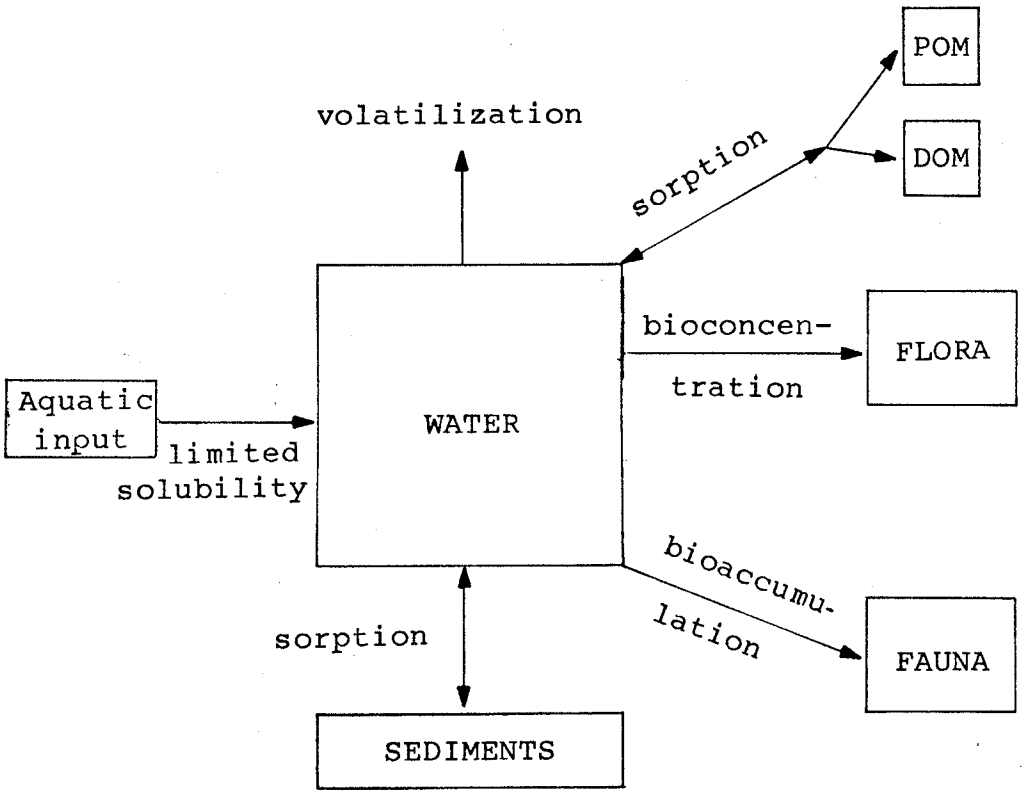


Figure 1. Operative Processes in the Pond System

Various physical constants have helped to identify the major processes likely to take place within the aquatic system. Table 3 lists various physical data and physical constant values of the non-ionic, O_8CDD.

Table 3

Physical data of O_8CDD

Molecular weight[c]		459.73 g/mol
Percent (%) by weight:	C	31.3
	O	7.0
	H	0
	Cl	61.7
Melting point[a]		325-326°C
Solubility (g/L) at 20-25°C:		
CH_3OH[a]		<0.1
Acetone		<0.5[a], 0.005[b]
DMSO[a]		0.5-2.0
$CHCl_3$[bc]		0.56
Benzene[b]		1
Toluene[b]		1.6
THF[a]		2.5-3.5
Xylene		3.6[b], 3.58[c]
Water solubility		
S_w (g/L) 20°C [d]		0.4×10^{-9}
Partition coefficients		
K_{ow} (20°C) [d]		8.3×10^{10}
K_{oc}		1.2×10^{8}[d], 8.3×10^{5}[e]
Vapor pressure		
V_p (atm) 20-25°C		1.78×10^{-10}[f], 2.37×10^{-10}[g]

[a]Spangler (1983), [b]Les Chlorophenols et leur impuretés dans l'environnement Canadien ESP (1981), [c]Esposito et al., (1980), [d]Webster (1984), [e]Muir et al., (1985), [f]Dobbs and Cull (1982), [g]US EPA (1978)

The chlorine content of O_8CDD differentiates O_8CDD from other dioxin isomers and gives it the greatest molecular weight of the series of isomers, with the chlorine atoms representing 62% of its molecular weight. Its solubility in organic solvents is not very large whereas its solubility in water (S_w) appears to be extremely small. Its vapor pressure (V_p) is also very low, possibly indicating its weak tendency to volatilize.

Octanol-water partition coefficient (K_{ow}) and organic carbon-water partition coefficient (K_{oc}) are key parameters in environmental fate studies of organic contaminants. Under environmental conditions, k_{ow} has been found to be related to water solubility, soil/sediment adsorption coefficients, and bioconcentration factors (BCF) for aquatic life (Lyman, 1982). k_{ow} values can have some environmentally significant implication as they could be thought to represent the tendency of a chemical to partition itself between an organic phase; fish, plants and/or sediments, and an aqueous phase.

O_8CDD has a high k_{ow} value (Table 3) and can be considered as a very hydrophobic chemical. It may well indicate that O_8CDD would preferentially partition itself into organic phases of an aquatic environment. Therefore, strong affinities for organic phases and low water solubility would appear to be characteristics of a chemical that is able to persist in aquatic ecosystems.

The bioaccumulation potential of highly hydrophobic chemicals such as O_8CDD may also be dependent on their molecular and physico-chemical properties (Bruggeman et al., 1984). It has been suggested that increasing molecular size and high lipophilicity might affect the linearity of the relationship between log K_{ow} and log BCF (Tulp and Hutzinger, 1978).

Muir et al., (1985b) reported BCF for O_8CDD much less than those predicted from correlations of k_{ow} and BCF. These correlations have generally been derived with compounds having log $k_{ow} < 6$ (Veith et al., 1979).

Bruggeman et al., (1984) studied the influence of size and hydrophobicity (lipophilicity) on the bioaccumulation of a series of xenobiotics in fish, via water and food. They selected the test chemicals so they possibly fall in the critical region where increasing lipophilicity does not necessarily result in higher bioaccumulation factors. Three high molecular size chemicals; hexabromobenzene, perchloroterphenyl and O_8CDD were not detected in living fish (detection limit 0.05 mg/Kg fish by GC-EC). However, they detected considerable quantities of these compounds in dead fish; apparent concentrations factors were 10^3 for O_8CDD and perchloroterphenyl and 10^4 for hexabromobenzene. The authors then concluded that for living fish, direct fish-water partitioning of O_8CDD did not occur. However, Stalling et al. (1983) and Muir et al. (1985b) report the partitioning process of O_8CDD into living fish.

Muir et al., (1985a) investigated the accumulation potential of four chlorinated dioxins (one of the tetra, penta, hexa, and hepta isomers) from water by rainbow trout fry (Salmo gairdneri) and fathead minnows (Pimephales promelas) for a 5 day exposure period. Low equilibrium BCF of the hexa and hepta isomer by both species (4232 for H_6CDD and 515 for H_7CDD in fathead minnows) were attributed to the high proportion of each chemical in water, apparently unavailable to the fish due to association with particulate and dissolved material.

Therefore, for highly hydrophobic chemicals such as O_8CDD, uptake by the fish may be more important via food than via water, as important concentrations could be found associated with detritus and microorganisms which form basis of aquatic food chains leading to fish.

Muir et al., (1985b) calculated BCF of O_8CDD by use of a two-compartment first order rate model, following a 5 day uptake and 24-48 day depuration period. Calculated BCF ranged from 85 to 2226 for O_8CDD in trout fry and minnows respectively. A dietary exposure of rainbow trout to O_8CDD resulted in a slow but steady accumulation of O_8CDD from food. These results may well indicate that, when describing the accumulation of O_8CDD in fish, a food chain transfer model might be more appropriate than equilibrium partitioning.

Combined with association to DOM and/or POM, which considerably reduce availability to the fish, uptake is possibly hindered by particular structural properties which may interfere with membrane transport. Bruggeman et al., (1984) suggested that from a mechanistic point of view, spatial dimensions (i.e. molecular thickness) rather than mass would interfere with membrane passage.

Therefore, more intrinsic properties of O_8CDD such as structure, molecular volume and surface area could possibly determine the potential of O_8CDD to partition itself into living organisms such as fish or plants.

5.0 Structural Characteristics

Prediction of the bioaccumulation of O_8CDD in fish, solely from its partition coefficients, may not be reliable since it has been suggested that molecular size and high lipophilicity affect the linearity of the relationship between log k_{ow} and log BCF (Tulp and Hutzinger, 1978). Bruggeman et al., 1984 ask to what extent a fish is comparable with a fat droplet in water. They suggest that uptake can possibly be hindered by particular structural properties interfering with membrane passage. In their opinion, spatial dimensions (i.e. molecular thickness) rather than mass would interfere with membrane passage.

McKinney and McConnell (1982) report that toxic structures (in the guinea pig) in the halogenated biphenyl, dibenzo-p-dioxin, dibenzofuran, and naphthalene compound classes have been identified and important structural requirement assessed.

5.1 Incumbrance Area. McKinney and McConnell (1982) defined the incumbrance area as the surface of the smallest rectangular envelope of a planar molecule, drawn proportionally to molecular dimensions.

Structural requirements for toxicity have been identified among dioxins, such as the incumbrance area of the toxic 2,3,7,8-T_4CDD, represented by the rectangular envelope of the isomer (McKinney and McConnell, 1982). Structural requirements found in 2,3,7,8-T_4CDD appear to have resulted in induction of cytochrome P-448 in rats and mice

and caused high toxicity in the guinea pig (McKinney and McConnell, 1982). Deviation from such rectangular arrangement has, as with 2,7-D_2CDD, significantly lowered the biological potency (Schewetz, 1973).

6.0 O_8CDD metabolism

In assessment of the persistence of a substance in the environment, the susceptibility of that substance to biodegradation is of primary concern (Esposito et al., 1980). The metabolism of PCDD's has not been investigated in detail, (Safe et al., 1983) but a number of in vivo studies have shown: (1) hydroxylation is favored at the 2,3,7,8-positions; (2) the rate of hydroxylation of specific substrates tends to decrease with increasing ring chlorination, although it is also dependent on the chlorine substitution pattern.

Tulp and Hutzinger (1978a) studied the rat metabolism of PCDD's. They observed hydroxylation of various isomers but no metabolites were found from O_8CDD.

Toxic effects of chemicals are usually observed in potential target organs such as the liver (Bickel et al., 1983). When no toxic effects are observed, it can be hypothesized that the chemical in question is being sequestered in adipose tissue away from potential target organs. However, if the adipose tissues are metabolized, stored material can be released. The chemical can then be transferred into lean tissues accompanied by increased

excretion via the feces. Williams et al., (1972) and Norback et al., (1975) studied the distribution and excretion of O_8CDD in the rat. The former group observed that a large quantity of O_8CDD was recovered in the feces while the latter reported that over 90% of the total dose was recovered in the feces as unabsorbed material. These results may well indicate that O_8CDD was being accumulated in adipose tissues but not retained since it had been excreted in important proportions in the feces as unabsorbed material. Adipose tissue storage of known lipophilic compounds does not seem then to be simply a matter of lipophilicity, and may be more complex than expected, especially for extremely lipophilic chemicals such as O_8CDD. Dobbs and Williams (1983) suggest that chemicals expected to have high octanol-water partition coefficients (k_{ow} $O_8CDD \sim 8 \times 10^{10}$ Table 3), on the basis of low water solubility relationships may also have low fat solubilities, which would have an influence on bioconcentration by limiting the amount of chemical that can be stored in fat deposits of organisms. Fat storage of lipophilic compounds may thus be influenced by structural features as well as by partition coefficients and lipophilicity.

III. EXPERIMENTAL

1.0 Apparatus and Chemicals

1.1 Chemicals. The ^{14}C labelled octachlorodibenzo-p-dioxin was purchased from Pathfinder Laboratories Inc. (St-Louis, Missouri). The isomer has a specific activity of 25.8 mCi/mmole. The standard was dissolved in THF and made up to 50 mL as a primary standard. All solvents used were Caledon pesticide grade distilled in glass (Caledon Ltd., Georgetown, Ontario). Liquid scintillation counting (LSC) cocktail: Scinti Verse, Universal LSC Cocktail, Fisher Brand Silica gel (60-200 mesh) reagent grade from Fisher. Magnesium sulfate ($MgSO_4$ anhydrous, Fisher Brand). Glass wool used was silane treated (Applied Science Laboratories Inc).

1.2 Apparatus. The HPLC apparatus used for ^{14}C-O_8CDD confirmation was a Waters Associates model 6000 A solvent delivery system, model 440 UV detector (254 nm), and a μBondapak C_{18} column.

The liquid scintillation counter was a Beckman LS-7500 instrument. Extraction efficiencies of sediment, fish, and plant samples were determined by combustion using a Packard 306 Oxidizer, which burned samples for total ^{14}C activity. Cellulose extraction thimbles (40 x 125mm) were purchased from Whatman, W & R Balston Ltd., England. Stainless steel

ball-mill tubes were constructed by J. Solomon (Solomon and Muir, 1981).

2.0 Experimental Area

2.1 Pond Construction and Description. The experimental site was located at the Glenlea Research Station, 20 km south of Winnipeg. Experimental ponds (3) constructed for a previous study (Corbet et al., 1983) were used; they were constructed in 1980 according to a method described by Madder (1978). The excavated holes (5.5m x 4.5m x 0.9m deep) were arranged as follows. The walls were hand smoothed and 5 cm of mortar sand was laid over the bottom and compacted. One layer of 10 mil polyethylene plastic was laid down and the bottom lined with 5 cm of sand. The walls were covered with two offset layers (5cm thick) of clay base sod. The bottom was then covered with a 10 cm layer of excavated earth. Dimensions of the empty ponds at the time of the study were: 4.9 m x 3.8 m x 0.5 m deep. Water was pumped into each pond to an estimated volume of 5 m^3. During the course of the experiment, water was pumped from a nearby dugout and was added when needed to maintain the water at the acceptable level of 0.38 m. Pond water characteristics at the time of ^{14}C-O_8CDD application are listed in Table 4.

Table 4

Water Quality Parameters

	Pond 2	Pond 5	Control
pH	7.1	7.5	7.2
Temperature (^{o}C)*	10.0	10.5	9.6
Suspended solids(mg/L)**	8.5	4.5	6.0

*Temperature measured at 5 cm below water surface.

**100 mL water sample filtered through 0.45 microns Millipore type filter paper.

2.2 Preliminary screening of ponds. In order to determine the level of contamination of the ponds from earlier work, duplicate samples of water (500 mL) and hydrosoil were taken from ponds 1, 2, 4, and 5 (Figure 2). Water samples were collected and extracted as described in sections 3.1 and 4.1. Hydrosoil samples were obtained by scraping the top 2 cm of the bottom of the pond. Each sample was vacuum filtered, using a Buchner funnel, air-dried, weighed accurately and combusted. Carbon-14 residues were analyzed by LSC. Results are presented in Table 5.

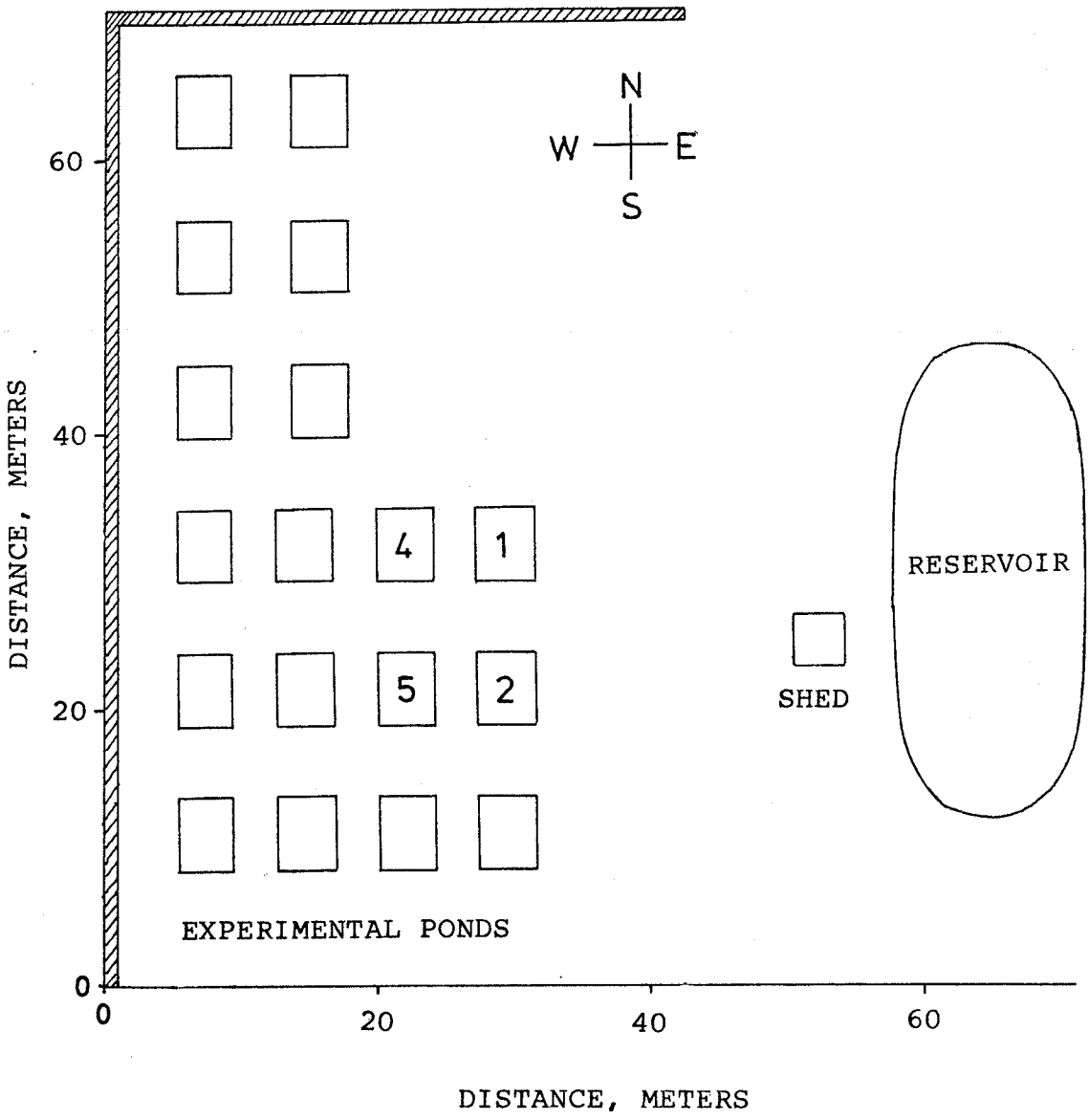


Figure 2. Pond site

Table 5

Level of Contamination of Ponds under Investigation

Pond	^{14}C Contamination of water (DPM)	^{14}C Contamination of sediments (DPM)
1	BKG	287±2
2	BKG	BKG
4	BKG	BKG
5	BKG	70±4

Background (BKG): 28.9 DPM

2.3 Pond Treatment. On August 17 1983, two ponds were treated with ^{14}C-O_8CDD. The water (5 m^3) was dosed with 1.7 mg ^{14}C-O_8CDD (pond 2) and 3.4 mg ^{14}C-O_8CDD (pond 5). Pond 2 was treated with 340 ng L^{-1} (33796 DPM L^{-1}), pond 5 with 680 ng L^{-1} (67592 DPM L^{-1}) and pond 4 was left untreated. The dioxin was coated on sodium chloride crystals following the procedure used to introduce DDE into a water column (Hamelink and Waybrant, 1976).

The dioxin was dissolved in THF (5.9 mg O_8CDD in 25 mL THF). 10 g sodium chloride were added to the solution and solvent slowly evaporated using a roto-evaporator. 5.76 g of salt (3.4 mg O_8CDD) was added to pond 5 (high treatment) and 2.88 g of salt (1.7 mg O_8CDD) was added to pond 2 (low treatment).

3.0 Sampling

Sampling of the various components of the ponds was started on August 17, 1983, and terminated on October 7. The sampling schedule is presented in Table 6.

Table 6

Sampling Schedule for ^{14}C-O_8CDD Treated Pond Compartments

Time	Air	Water	Sediments	Vegetation	Fish
0	-	-	-	-	-
0.2h	Sampled every 2 h for 36 h.	X		X	X
2h		X	X	X	X
4h		X			
8h		X			
12h		X	X		X
24h		X	X	X	X
2d		X	X	X	X
4d		X	X	X	X
8d		X	X	X	X
16d		X	X	X	
24d		X	X	X	
34d		X	X	X	
51d		X	X	X	
382d			X		
652d			X		

3.1 Water. Subsurface water (5 cm below surface) was collected (900 mL) by submersing a one-litre bottle in the water column. The lid of the bottle had two pieces of tubing, one short piece to let the water in and a long piece to allow the air to escape from the bottle. Dichloromethane (50 mL) was added to each sample immediately after their collection. Samples were contained in clean 4-L solvent bottles and stored at 4°C while awaiting analysis.

3.2 Air. Air sampling was done by means of a series of three glass tulip funnels (5 cm x 10 cm) inverted and located at 3 different heights above the water surface: 10, 20 and 50 cm. Each funnel was provided with a polyurethane foam plug and air was drawn through each foam, by means of an attached vacuum pump, at a flow rate of 10 L min^{-1}. At every sampling time, (every two hours for 36 hours), foams were replaced with new ones, stored in jars and transported immediately to the laboratory for analysis. Wind speeds were recorded at heights of 10 and 50 cm by means of three-cup anemometers.

3.3 Sediments. Thirty days prior to the ^{14}C-O_8CDD application to the ponds, metal tins (12 cm i.d. by 7 cm deep) equipped with wire handles were filled with sediment from an unused pond, and placed on patio blocks that had previously been put on the bottom of each pond. A total of 40 tins was placed in each pond. At each sampling time, lids were

put on the tins while still on the bottom of the pond and tins were then lifted out the water. The top 2.0 cm of sediment was collected, put in jars and stored at -40°C while awaiting analysis.

3.4 Fathead minnows. Two weeks in advance of treatment, 100 minnows (Pimephales promelas) were put in each pond. Sampling of the fish was conducted using a minnow trap from which two or three minnows were taken at each sampling time. The fish were stored in jars at -40°C while awaiting analysis.

3.5 Vegetation. Rooted pondweed (Potamogeton berchtoldi) was sampled by hand, pulling from the floating part of the plant. Non-rooted macrophyte (Lemna minor) was sampled from the surface by means of a dip screen. Samples were stored in jars at -40°C while awaiting analysis.

4.0 Extraction

4.1 Water. Extraction of ^{14}C-O_8CDD from the water was initiated in the field by the addition of dichloromethane (50 mL) immediately after the collection of the nonfiltered water sample. In the laboratory, the water sample was transferred to a 2 L separatory funnel and further extracted with dichloromethane (3 x 30mL). The organic extract (140 mL) was dried on a magnesium sulfate column (10 g), reduced to 2 mL (roto-evaporator) and analyzed by LSC.

4.2 Polyurethane Foam Plugs. Foams were collected and transported to the lab immediately after each sampling time. Foams were soxhlet extracted for 2 h with hexane (200 mL). The extract was reduced (roto-evaporator) and transferred into screw cap test tubes. The extract was then evaporated to near dryness by blowing dry air onto it, made up to 1.00 mL in hexane and aliquots analyzed by LSC.

4.3 Sediments. The thawed sample was homogenized by stirring, weighed into a cellulose extraction thimble (25 to 30 g wet weight), covered with glass wool and soxhlet extracted for 12 h with THF (300 mL). The sample was then further extracted for 6 h with fresh THF (300 mL). Each of the 300 mL extract was treated separately as described below.

Each extract was reduced in volume by roto-evaporation to approximately 10 mL and then made up to 100 mL with water. The aqueous phase was partitioned with benzene (3 x 50mL). The 150 mL benzene extract was reduced (roto-evaporator) to approximately 2 mL and further reduced to near dryness under a stream of dry nitrogen. The residue was made up to 1.00 mL in hexane and cleaned up as follows.

A pasteur pipette fitted with a glass wool pledget was packed from bottom to top in the following manner: 0.8 g non-treated silica (60-200 mesh) and 0.8 g silica impregnated with concentrated H_2SO_4 (44% by weight). The column was prewetted with hexane and eluted with hexane (5mL). The

eluent was collected in LSC vials and total ^{14}C activity determined by LSC.

4.4 Fathead minnows. One to three minnows were extracted by the ball-mill technique. Minnows (3.0-5.0 g total) were thawed, blotted dry, sliced into pieces with a scalpel and placed in a stainless steel tube along with 5.0 g magnesium sulfate and two stainless steel balls. The fish tissue was extracted with THF (20 mL) on a wrist-action shaker for 45 min. The tube was centrifuged at 1000 rpm for 15 min., the supernatant pipetted off, and the procedure was repeated twice. The THF extract was reduced to a small volume (roto-evaparator) and evaporated down to near dryness under a stream of dry nitrogen. The residue was made up to 1.00 mL in benzene and aliquots cleaned up as follows.

A microcolumn such as those described in section 4.3 was used. The column was prewetted with benzene and the elution done with benzene (5 mL). The eluent was collected in LSC vials and analyzed by LSC.

4.5 Vegetation. Duckweed and rooted vegetation samples were extracted by the ball-mill extraction technique. The plant material was thawed, blotted dry, weighed (5.0 to 6.0 g) and put in stainless steel tube with magnesium sulfate (5.0 g) and two stainless steel balls. The plant material was extracted with THF (20 mL) on a wrist-action shaker for 50 min. The tube was centrifuged for 15 min. at 1000 rpm and

the supernatant pipetted off. The procedure was repeated twice. The plant extract (50 mL) was concentrated to a small volume (roto-evaporator) and further reduced to near dryness under a stream of dry nitrogen. The residue was made up to 1.00 mL in hexane and cleaned up as follows.

A chromatography column (150 mm x 15 mm i.d.), fitted with a glass wool pledget, was packed with 10 g deactivated silica (60-200 mesh) for duckweed samples and 12 g deactivated silica for rooted vegetation samples. Each column was prewetted with hexane and eluted with hexane (20 mL); 3-mL fractions were collected and analyzed by LSC.

5.0 Analytical Methods

5.1 High-pressure Liquid Chromatography. Environmental samples were analyzed for ^{14}C-O_8CDD by HPLC. The solvent flow rate was 0.8 mL min^{-1}, the mobile phase consisting of neat methanol. Elution time was 8.2 min. After sample injection, fractions (2 mL) were collected in scintillation vials, starting at 5 min., (total volume collected 20 mL) for LSC analysis.

5.2 Combustion. Extraction efficiencies of solid samples were determined by combustion on a Packard 306 oxidizer. Duplicate samples (0.3 to 0.5 g) were oxidized and $^{14}CO_2$ was trapped in CO_2-M-Met, diluted with PCS-xylene (2:1) and followed by LSC.

6.0 Calculation of Structural Characteristics

6.1 Incumbrance Area. Bond lengths and angles of the hexa isomer 1,2,3,7,8,9-H_6CDD were determined by X-ray diffraction and were reported by Cantrell et al. (1969). Bond lengths and angles of O_8CDD and 1,3,6,8-T_4CDD were approximated from his data. To the bond lengths, van der Waals radius of chlorine atoms (r= 1.8 Å) were added and the smallest rectangular envelope drawn.

6.2 Molecular Volume. The van der Waals volume (v_w) of dioxin congeners was calculated following a method described by Edward (1970). The v_w of O_8CDD and other dioxins was obtained by the addition of atomic increments of Table 7. The idea is to dissect the molecular volume into contributions from individual atomic increments (Edward, 1970).

Table 7

Van der Waals Increments of Atoms (Edward, 1970)

Atom	Increment ($Å^3$)
C=	8.1
-O-	6.2
H-[a]	5.2
Cl-	19.8

[a]Aromatic

7.0 Compartment Size Estimation

7.1 Water Compartment. Volume of the water was estimated to be 4.9 m^3. Volume was estimated by using the size of the ponds (4 x 5 x 0.5 m) and the slope of the walls, as reported by Corbet et al. (1983). Muir et al. (1985c) used identical ponds and estimated the volume at 5.3 m^3.

7.2 Sediment Compartment. Bottom surface area of the ponds estimated to be 11 m^2 including 60 cm sides. Knowing the average sample depth (2 cm) and the density of the sediment material (0.8 g/cm^3), we estimated the weight of the sediment bed (top 2 cm) at 172000 g.

7.3 Vegetal Matter Compartment. Wetzel, (1983) reports pondweed bioamass estimations of various lakes which were approximated to our pond characteristics.

IV RESULTS AND DISCUSSION

1.0 Air Compartment

Volatile loss of the high molecular weight and low vapor pressure octachloro isomer was not expected to be very important overall, with most of the volatilization being expected to take place early in the experiment. Corbet et al. (1983) showed that under similar field conditions, the tetra isomer 1,3,6,8-T_4CDD volatilized rapidly from the water, with the greatest loss occurring within the first 24 h of their study. Extraction efficiency for 1,3,6,8-T_4CDD from foams was 80 % and over (Corbet et al., 1983).

It was thus decided to sample the air over the treated pond intensively during that period, more specifically at every 2 h, hoping to observe a trend of some sort.

The dioxin was applied to the water coated onto sodium chloride crystals, a method that ensured the total and rapid dispersion of the octa isomer into the water column. Corbet et al. (1983) noted some important variability in water concentrations early in his experiment, variabilities that were believed to be due to incomplete mixing of 1,3,6,8-T_4CDD in the pond at the time of treatment, since it was applied subsurface as a solution in acetone. The water temperature was 10.5 ^{o}C, the salt dissolved rapidly and completely at that temperature (NaCl solubility in water (0 ^{o}C) 35.7 gm/L).

Overall volatilization of O_8CDD was found to be almost non-existent. The method itself is more qualitative and it shows, at best, whether volatilization is taking place or not. A great number of climatic conditions (Figure 3) may have affected the volatilization process in ways not yet completely understood.

Results obtained show no noticeable trend of the concentration of released dioxin in the air over the ponds during the 36 h experiment; the levels fluctuated quite widely indicating the uncertainties involved in quantitating environmental chemicals at these very low vapor concentrations. Figure 4 presents volatile losses of ^{14}C-O_8CDD above each treated pond, each bar representing losses at three different heights. The low treatment pond had two important volatilization periods, 1130 in the first day (August 17) and 1330 in the second day. Two important volatilization periods were also identified above the high treatment pond, although observed at different times of the day. Concentration of O_8CDD in air at 1930 on day 1 was 22 pg/L and 3 pg/L at 1730 on day 2 (detection limits of approximately 1 pg/L).

Figure 4 also presents wind speed variations throughout the 36 h experiment. The two major volatilization peaks were observed above the low treatment pond at times when low wind speeds were recorded. Wind speed measurements were taken above the low treatment pond only. Because of physical differences viz., vegetation cover between the two

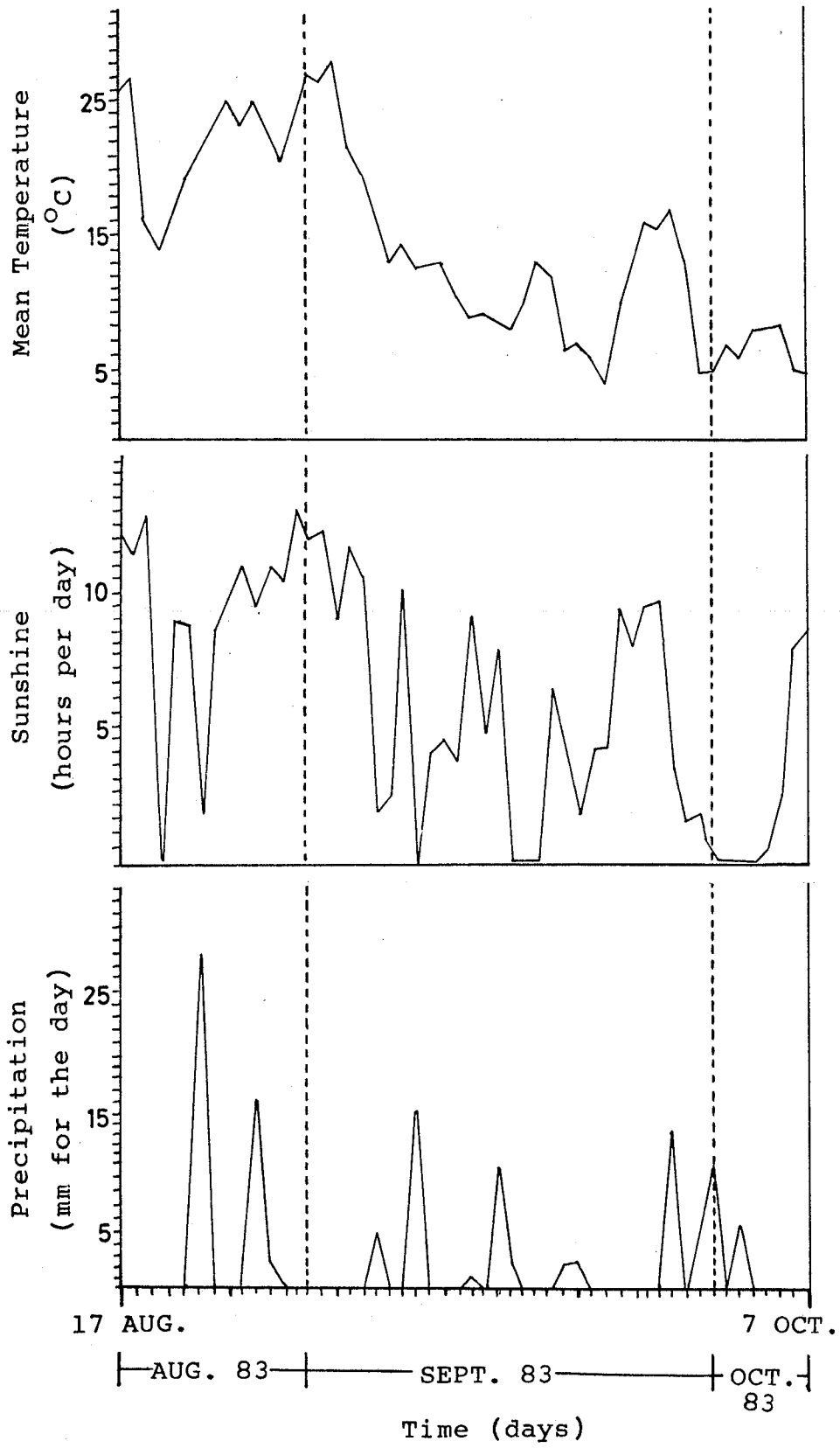


Figure 3. Meteorological Data

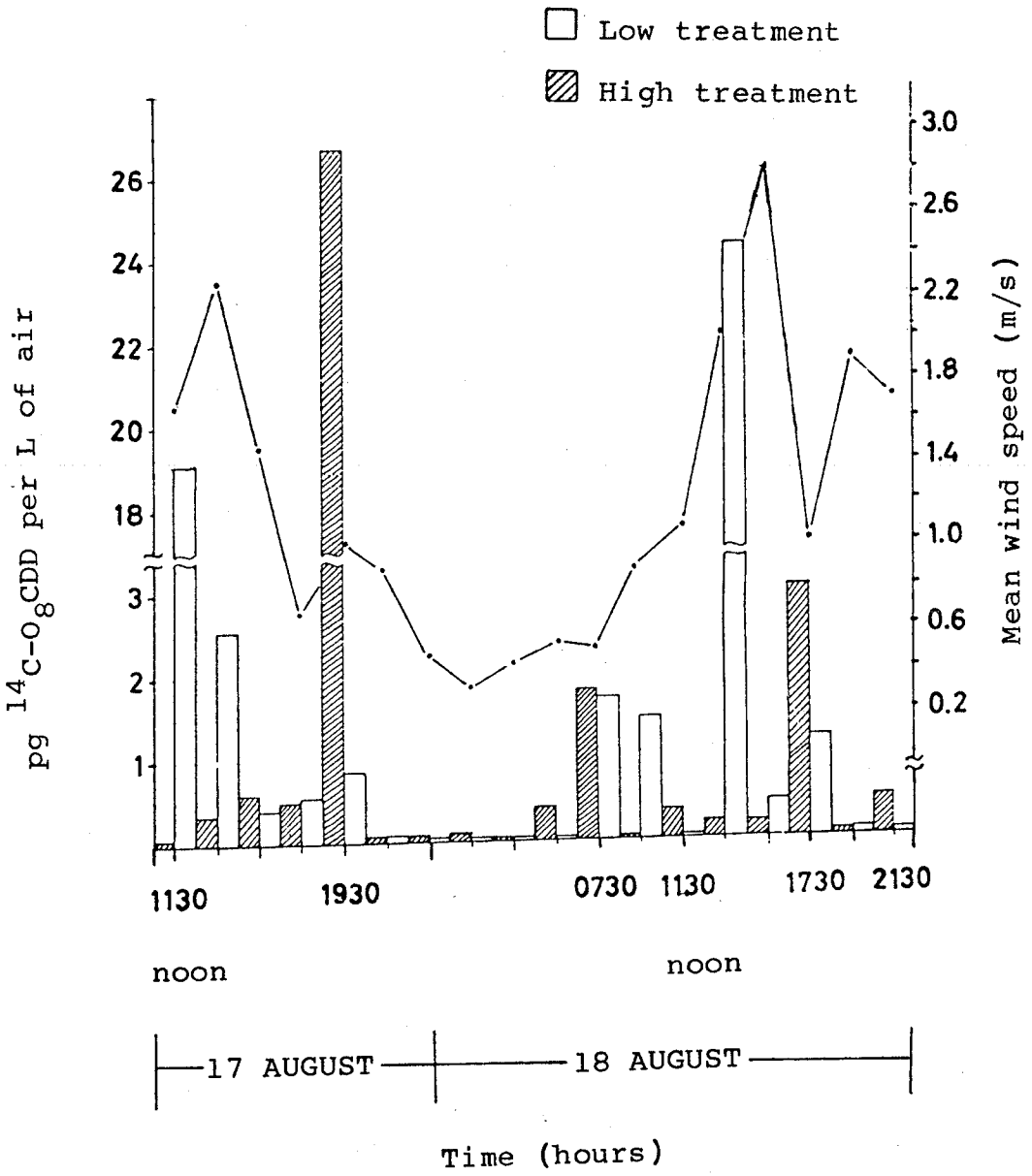


Figure 4. Volatile Losses from ^{14}C-O_8CDD Treated Ponds

ponds, these wind speed values are valid only in the case of the low treatment pond.

1.1 Pond Physical Description and Meteorological Parameters.

Wind speed data and pond water temperatures were measured directly on the site. Other weather measurements, viz., air temperature, sunshine record and precipitation were recorded and obtained at the Glenlea Research Station climatological lab, located within 1 km of the pond site.

Various physical and meteorological factors are relevant and probably had some effect on the volatilization pattern observed. Physical characteristics of the ponds themselves give some valuable information. Figure 5 presents a schematic description of the two treated ponds as they were on the morning of August 17. The low treatment pond was heavily colonized with cattails, mostly on the south side, where plants reached approximately 1.0 m from the surface of the water. This vegetation wall offered good protection against the prevailing southwest wind blowing that day. The high treatment pond contained no cattails but was covered, over approximately 75% of its surface, by the floating macrophyte Lemna minor.

Figure 5 demonstrates that air flow above each pond may have been different. The vegetation in the low treatment pond would have slowed the motion of the air and also caused the wind to deviate markedly from its initial pattern. The roughness of the terrain determines the angle at which the

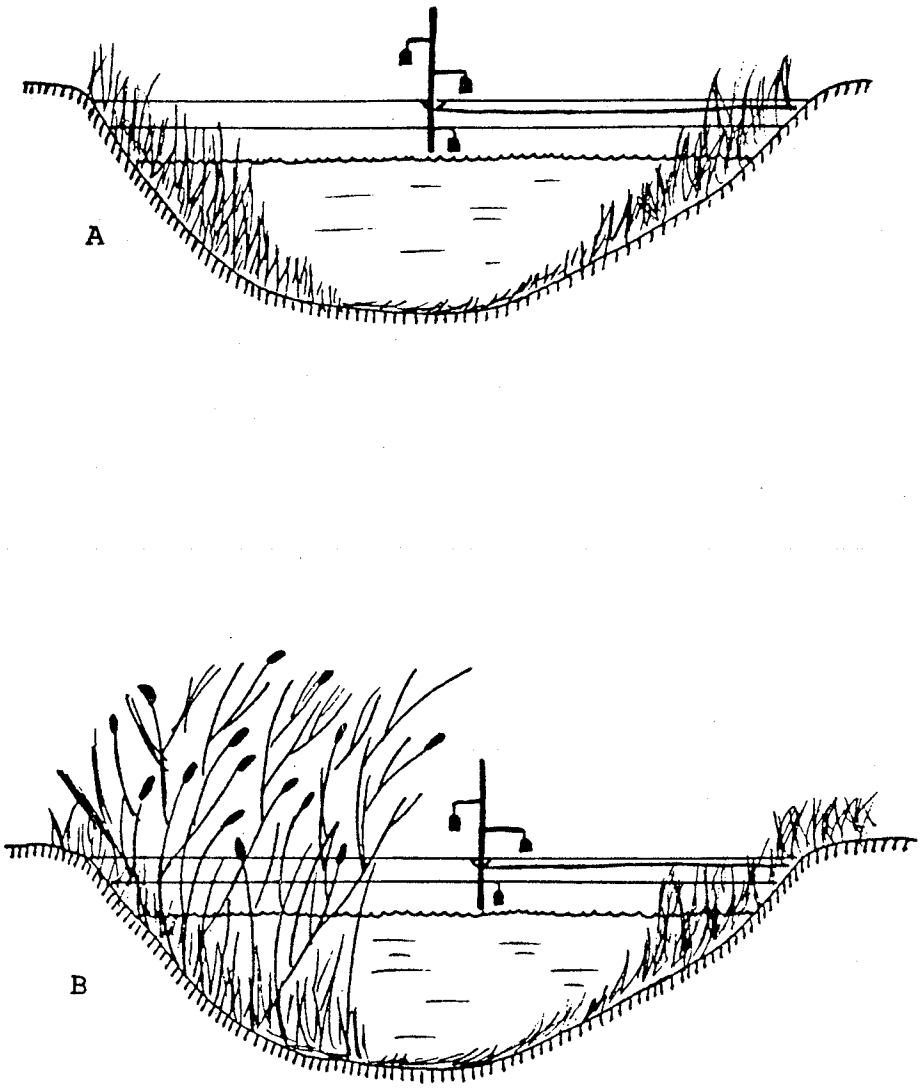


Figure 5. Pond Physical Characteristics: High Treatment (A) Low Treatment (B).

air will flow as well as the speed at which it will move (Lutgens and Tarbuck, 1979). The resulting effect on the dioxin concentration in the air above that pond was likely that the dioxin concentration was greater as opposed to diluted above the high treatment pond, where no such vegetation was present.

Combined with physical characteristics of the ponds, the stability of the air above the pond very likely affected the fate of the released dioxin and subsequently, the concentration variations observed. The degree of atmospheric turbulence depends primarily on the stability of the air and the properties of the wind. The stability of air is directly affected by its temperature, which in turn may affect the diffusion rates of pollutants. Ordinarily, the air tends to be unstable when incoming solar radiation is greatest; intense solar heating causes the lower air to rise. This upward movement of air, combined with light wind conditions, enhances atmospheric turbulence. Such conditions of high turbulence were present at certain times of the day, mostly before and shortly after noon. These conditions would have occurred above the low treatment pond, at 1130 on day 1 when O_8CDD concentration in the air was 16 pg/L and at 1330 on day 2, when O_8CDD concentration in the air was 20 pg/L. At these two times, the highest concentrations were observed at 5 cm above water surface, suggesting that upward movement of warm air was possibly not important.

In the case of the high treatment pond, volatilization appeared more important during the evenings, at 1930 on day 1 and at 1730 on day 2. At these two particular times, outgoing radiation exceeded incoming radiation as a result of the water emitting heat stored during the day. This leads to the cooling of a shallow layer of air near the surface and subsequently, the formation of a temperature inversion. Air near the surface is cooler and heavier than the air aloft, therefore, little vertical mixing takes place between layers. Since the dioxin is coming from below, the temperature inversion would confine it to the lowermost layer, where it would continually increase in concentration. However, certain pockets of air may have been warmed more than the surrounding air, and subsequently moved upward, along with the released dioxin, perhaps explaining why the concentration of O_8CDD in air was 22 pg/L at 50 cm on day 1, a maximum during that day.

At night, low-level temperature inversions tend to prevail, especially following hot summer days, where temperature differences from day to night are very high (Lutgens and Tarbuck, 1979). In our study, the air temperature reached 35^{o}C on the day of August 17 and dropped to 10.5^{o}C overnight. Whenever such conditions are accompanied by light winds, diffusion rates are particularly small and pollutants emitted accumulate in the inversion layer (Battan, 1979). Dioxin emission was effectively very poor on the night of August 17.

1.2 Volatilization Rate. The most widely accepted model to describe volatilization is called the two-resistance model or two-film theory. This theory assumes a two-layer boundary at the air-water interface (Figure 6), which offers most of the resistance to diffusion of the chemical from water to air or air to water.

The value of Henry's constant indicates the direction of transfer, i.e., whether volatilization of the chemical is controlled by diffusion in the liquid phase or the gas phase, thus, determining where a chemical would tend to go to accumulate in aquatic systems, assuming no other input or removal processes. Henry's constant can be expressed as the ratio of vapor pressure to water solubility (Mackay et al., 1979).

$$H = V_p/S_w \quad \text{Pa } m^3/\text{mol}$$

Henry's constant can be calculated from vapor pressure and aqueous solubility determined experimentally. Such calculations may be suspect, especially in the case of hydrophobic chemicals such as O_8CDD, since numerous difficulties may be encountered upon determination of their V_p and S_w (Mackay, 1980). The calculated value of Henry's constant for O_8CDD is approximately 2×10^4 Pa m^3/mol, indicating that O_8CDD would tend to accumulate in the air above the water, from which it would have been present in the truly dissolved state.

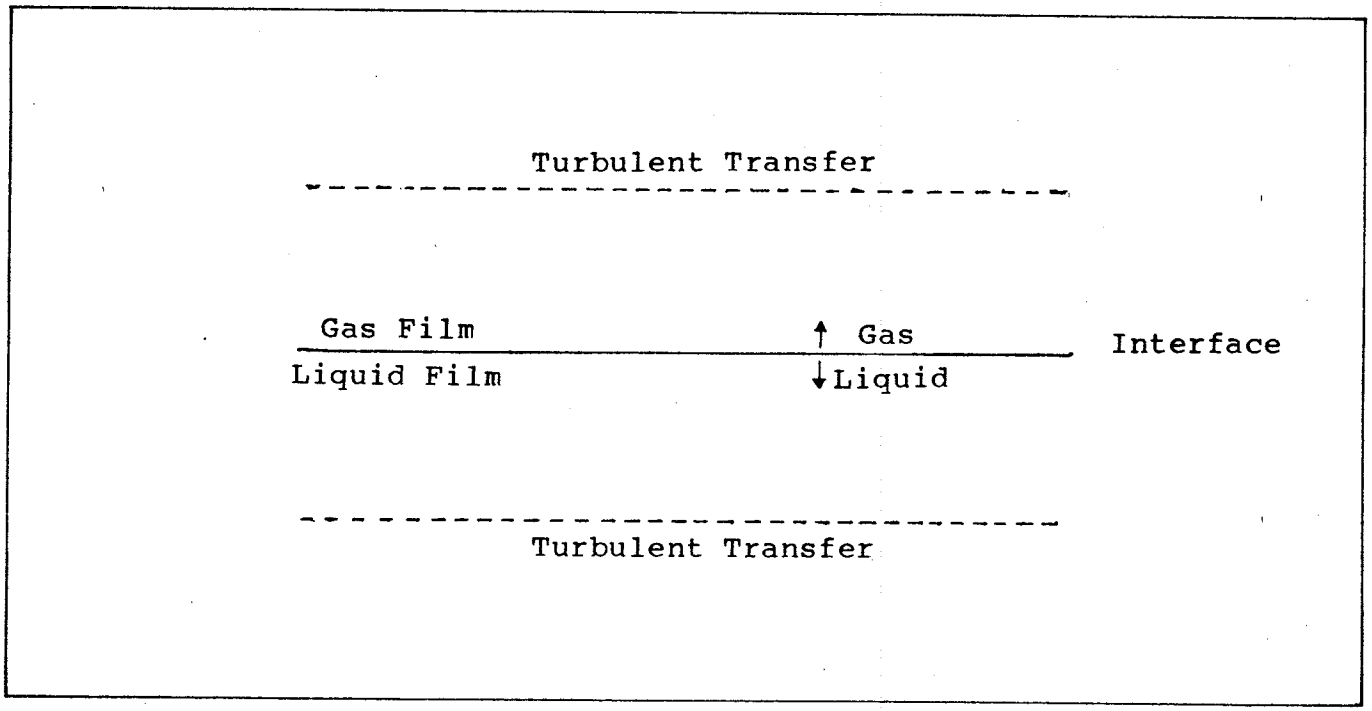


Figure 6. Two-layer Model of a Gas-liquid Interface (Liss and Slater, 1974).

A second approach is to use the aerodynamic equation (Thibodeaux, 1979), in which concentration gradients (ng/m^3), temperature gradients and wind speed gradients are used to calculate the flux (ng/m^2- day) of a chemical from a surface. In our study, concentration gradients were not observed in samples taken at the three heights investigated. As a result, our set of data would not seem to be adequate for such calculations.

Under environmental conditions, the behaviour of chemicals is rendered complex by a number of factors. As a result, equilibrium at environmental interfaces is not attained. Subsequently, the prediction of evaporation rates becomes extremely difficult because there is often uncertainty about the exact physical state of the volatilizing solute (Mackay, 1980). The two-film theory and the aerodynamic approach assume that the chemical concentration measured in water is truly dissolved and not sorbed to suspended solids or dissolved organics. Thus, to correctly estimate volatilization rates of chemicals, the fraction in true solution is required. The role of sorption in reducing volatilization is a recognized fact, although it has been shown to be very difficult to quantify (Mackay at al., 1979).

2.0 Water Compartment

Within the first 8 d of our study, O_8CDD disappeared rapidly from the water column, at a calculated rate (first order model; NRCC, 1981) of $1.75 \times 10^{-2} h^{-1}$ ($r^2 = 0.76$) in the low treatment pond and $1.55 \times 10^{-2} h^{-1}$ ($r^2 = 0.69$) in the high treatment pond (Figure 7). Equilibrium was apparently reached on day 8 in both ponds, at a time when no further significant variations in O_8CDD concentration were observed. Beyond day 8, dissipation from the water to other pond compartments had somewhat stabilized, as it can be seen by the slow dissipation rates of $1.68 \times 10^{-3} h^{-1}$ ($r^2 = 0.90$) in the low treatment and $8.55 \times 10^{-4} h^{-1}$ ($r^2 = 0.89$) in the high treatment pond.

Water samples were not filtered prior to extraction and as a result, concentrations are somewhat overestimated, as they remain well above solubility of O_8CDD in water (0.4 ng/L). It appears that an important proportion of O_8CDD present in the water would be sorbed to POM and/or DOM found in the water of the ponds.

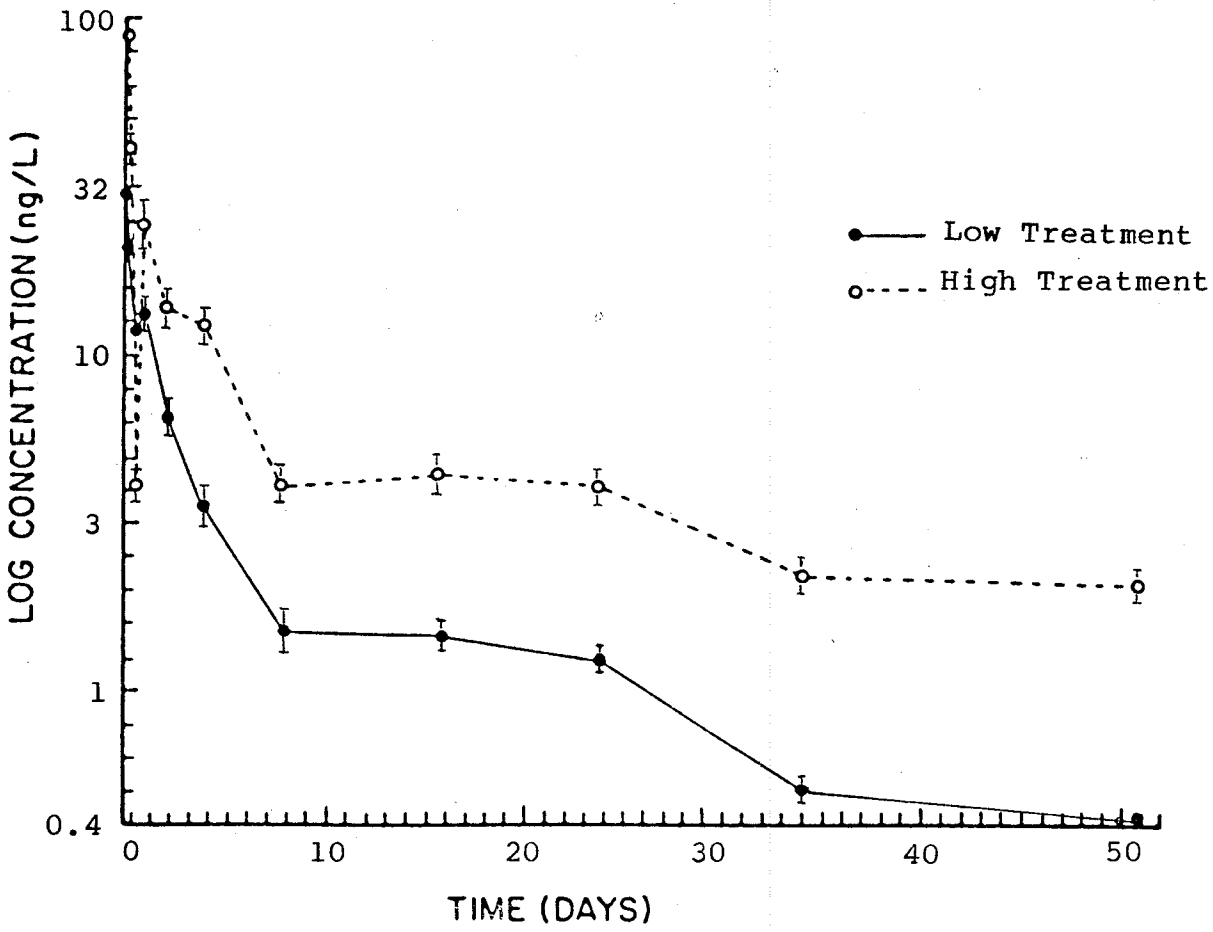


Figure 7. Concentrations of O_8CDD in Water of the Ponds
Mean bars represent range

3.0 Fish

Accumulation of ^{14}C-O_8CDD in the fathead minnows (Pimephales promelas) was detectable as early as 15 min. post-treatment, when 2.5 ng/g and 3.4 ng/g of the octa isomer were present in the fish from the low and high treatment ponds, respectively. Highest concentrations of ^{14}C-O_8CDD in the fish were found on day 1, when the fish had concentrated as much as 6.9 ng/g in the low treatment and 7.5 ng/g in the high treatment pond. Concentrations are reported on a wet weight basis, as total ^{14}C in a whole fish is the sum of extractable and unextractable radioactivity. The concentrations measured on day 1 represent an estimated 13 ng per fish, for a fish weighing an average of 1.8 g.

Beyond day 1, the concentration of ^{14}C in the fish did not increase; concentrations decreased consistently from then on (Figure 8). Lack of available fish made sampling impossible beyond day 8.

Concentration factor, CF, defined as the ratio of the concentration of a given chemical in the fish to the concentration in the surrounding water, could be calculated from field data. Results are presented in Table 8. Calculated CF were relatively lower within the first day probably because of the high O_8CDD concentrations in water observed at that time. The fish did not seem to be capable of accumulating O_8CDD from the water to the extent indicated by k_{ow} data (Sarna et al., 1984), as calculated CF's were never higher than 1300.

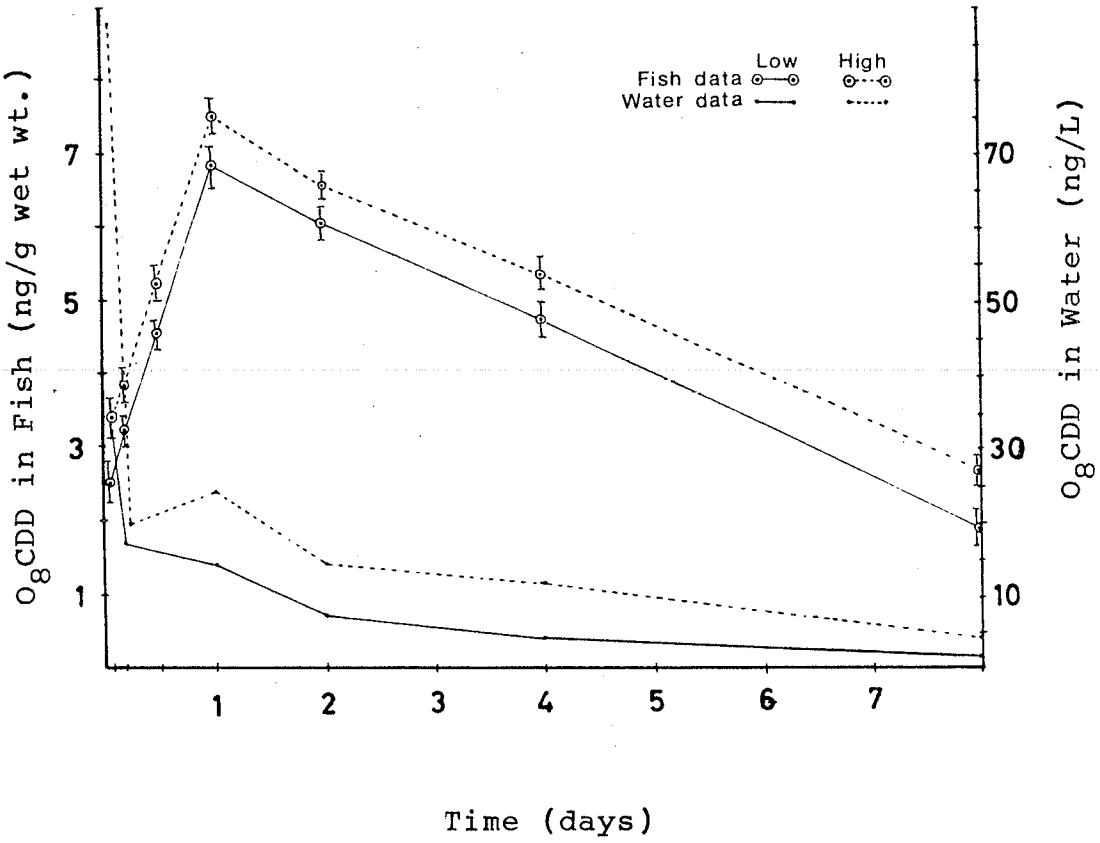


Figure 8. Accumulation of O_8CDD in Fish

Mean bars represent range

Table 8
Concentration of ^{14}C-O_8CDD* in fish

Time (days)	Low Treatment Concentration			High Treatment Concentration		
	Water ng/L	Fish ng/g	CF	Water ng/L	Fish ng/g	CF
0	-	-	-	-	-	-
0.01	32.3	2.5	78	89.2	3.4	78
0.08	17.4	3.2	188	19.3	3.8	190
0.5	12.5	4.5	346	4.3	5.2	1300
1	14.1	6.9	492	24.2	7.5	312
2	6.9	6.0	857	14.1	6.6	471
4	3.7	4.8	1200	11.9	5.4	450
8	1.5	1.9	950	4.2	2.7	675

*All ^{14}C-labelled material proved to be ^{14}C-O_8CDD by HPLC.

Muir et al. (1985b) calculated very low BCF's (100-300) following a 5 day exposure of fathead minnows and rainbow trouts to flowing aqueous solutions of ^{14}C-O_8CDD. These lower than predicted BCF's for O_8CDD appeared to be the result of low bioavailability due to association of ^{14}C-O_8CDD with suspended and dissolved organic carbon (DOC). In another study, Muir et al. (1985) investigated the effects of sorption of O_8CDD to DOC on its bioavailability from water. They found that centrifugation (20000 x g) of the water removed as much as 85% of ^{14}C-O_8CDD from solution, indicating its association to suspended solids. They then took the centrifuged solution and estimated that O_8CDD could associate to DOC in a very high

proportion, with a DOC partition coefficient in the order of 8.3×10^5. These results may well explain why we obtained low CF's (38-1300) calculated from total concentrations of O_8CDD in water i.e. the free and adsorbed fractions.

The bioavailability of a chemical from water to the fish is limited by the maximum concentration in solution, or by its water solubility (Spacie and Hamelink, 1982). Thus, the higher concentrations of O_8CDD found in fish from the high treatment, compared to the low treatment pond, would not result from a greater bioavailability of O_8CDD to the fish, since, throughout the experiment, concentrations of O_8CDD in the water of each pond were above the water solubility of O_8CDD (0.4 ng/L at 20^oC). The parallelism of the curves in Figure 8 could well result from this constant and equivalent bioavailability of O_8CDD to the fish. However, the lower concentrations of O_8CDD in the fish from the low treatment probably resulted from a lower bioavailability of O_8CDD in the fish, which was more than likely reduced because of a greater concentration of suspended solids, 8.5 mg/L, compared to 4.5 mg/L in the high treatment pond (Table 4). Landrum et al. (1984) have suggested that particulate organic matter (POM) and DOM-bound chemicals are unavailable to the fish.

Bruggeman et al. (1984) studied the influence of molecular size and hydrophobicity on bioaccumulation kinetics of various chemicals in guppies. They found that ^{14}C-O_8CDD was not accumulated by living fish and attributed their

results to possible structural or physico-chemical properties interfering with membrane transport. Muir et al. (1985) observed upon a 5 day exposure experiment that O_8CDD had very low rates of uptake and unexpectedly rapid rates of elimination (5-13 day half-life). Such results correlate well with my observations that the fish did not appear to accumulate the octa isomer beyond day 1, as the concentration of O_8CDD in the fish decreased constantly from that day on. Muir et al. (1985) suggested that O_8CDD might have been adsorbed to epithelial cells, as opposed to being absorbed, and subsequently lost due to natural turnover.

4.0 Vegetation

Rooted (Potamogeton berchtoldi) and floating (Lemna minor) (Figure 9) vegetation samples were analyzed for total ^{14}C activity by LSC. All ^{14}C was shown to be ^{14}C-O_8CDD, qualitatively, by HPLC retention times.

Floating vegetation (duckweed) appeared to bioconcentrate ^{14}C-O_8CDD quite significantly in the low treatment pond, with CF maxima in the order of 6000 and 7500 on day 8 and 16, respectively. In the high treatment pond, duckweed concentrated much less ^{14}C-O_8CDD, with highest calculated CF of 583 and 464, measured on day 1 and 2, respectively (Table 9). This difference may be explained by the presence of more floating vegetaion in the high treatment than in the low treatment pond, resulting in a somewhat greater dilution of the dioxin in the floating vegetation present.

Rooted vegetation represented the bulk of the vegetation in the high treatment pond, with an estimated biomass at seasonal maximum of 0.9 to 1.1 Kg wet weight per m^3 (Figure 10). In the low treatment pond, the emergent macrophyte (i.e. cattail, Typha) and rooted vegetation (Potamogeton berchtoldi) represented the bulk of the vegetation, with estimated biomass at seasonal maximum of 2.0 to 3.0 Kg wet weight per m^3 and 0.9 to 1.1 Kg wet weight per m^3, respectively. In spite of the presence of such an important volume of vegetal matter, the rooted vegetation was not able to concentrate the label very well

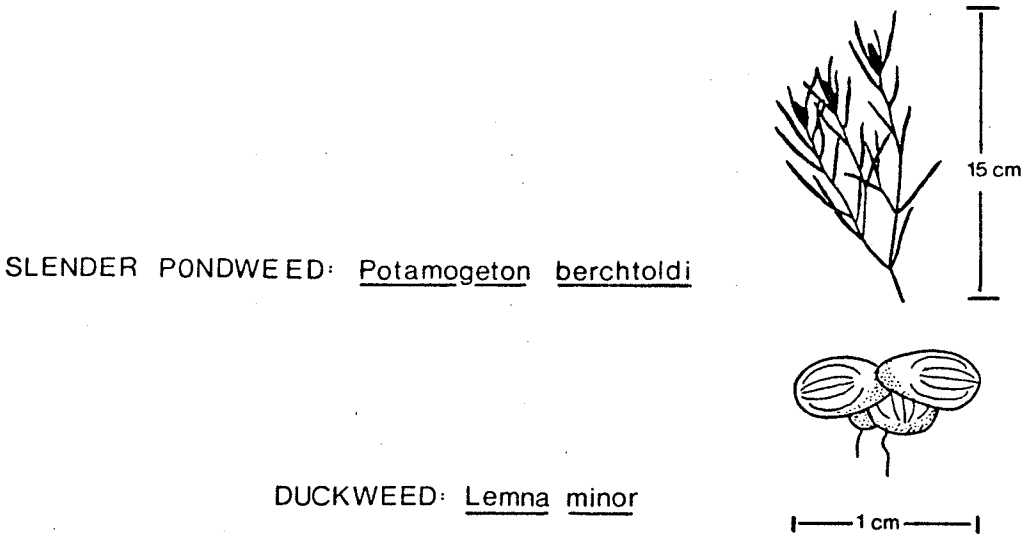
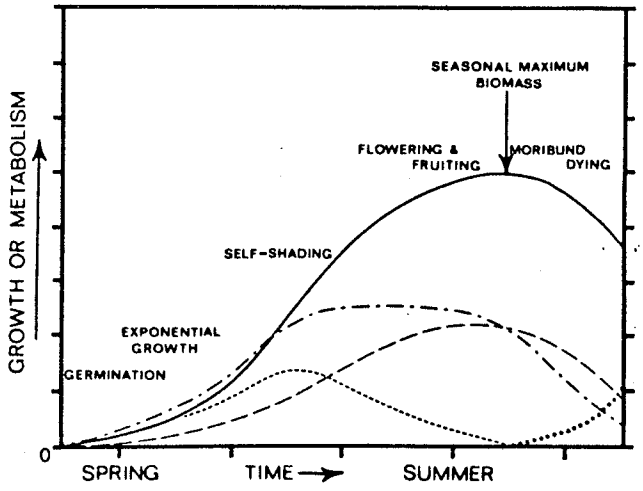


Figure 9 . Aquatic Macrophytes Sampled from the Ponds



——— = total biomass; — · — · = current gross productivity; · · · · = current net productivity; - - - = current respiration rate; · · · · · = death losses.

Figure 10. Generalized Growth and Metabolic Patterns for a Typical Annual Macrophyte (Wetzel, 1983).

Table 9

Concentration of O_8CDD in Vegetation

Time (days)	Low treatment					High treatment				
	O_8CDD in water ng/L	O_8CDD in plants *** ng/g wet wt. Floating	CF	Rooted	CF	O_8CDD in water ng/L	O_8CDD in plants ng/g wet wt. Floating	CF	Rooted	CF
0	ND	ND	-	ND	-	-	ND	-	ND	-
0.01	32.3	4.9 ± 0.3****	153	0.7 ± 0.1	22	89.2	1.5*	17	0.6 ± 0.1	7
0.08	17.4	5.4 ± 1.3	318	lost	-	19.3	3.5 ± 0.4	175	1.5 ± 0.9	75
1	14.1	3.11 ± 0.04	222	1.21 ± 0.06	86	24.2	14 ± 5	583	2.2 ± 0.4	92
2	6.9	1.4 ± 0.4	200	2.0 ± 0.2	286	14.1	6.5 ± 0.9	464	3.4 ± 0.7	243
4	1.5	1.4	350	2.14 ± 0.06	535	11.9	2.8 ± 0.2	233	4.3 ± 0.3	358
8	1.4	12 ± 2	6000	1.1 ± 0.1	550	4.2	0.46 ± 0.04	115	1.72 ± 0.04	430
16	1.3	7.5 ± 0.6	7500	0.8 ± 0.1	800	4.3	0.35 ± 0.01	88	0.05**	125
24	0.5	0.12 ± 0.05	120	0.26 ± 0.02	260	4.1	0.06 ± 0.01	15	0.27 ± 0.03	68
34	0.1	0.15 ± 0.07	150	0.2**	200	2.2	0.1**	50	0.3**	150
51	ND	ND	-	0.2**	-	1.9	ND	-	0.2**	100

**** Concentration ± range (Duplicates)

*** Radioactivity shown to be ^{14}C-O_8CDD by HPLC

** One of the two duplicates below detection limits

* One of the duplicates was lost

ND: None detected

in either pond, with highest CF's being 800 in the low treatment (day 16) and 430 in the high treatment pond (day 8) (Table 9). Corbet et al. (1983) also observed that rooted vegetation did not accumulate 1,3,6,8-T_4CDD to the magnitude found in the floating vegetation.

Overall, both floating and rooted vegetation bioaccumulated more ^{14}C-O_8CDD than the fish, an observation somewhat more evident in the low treatment than in the high treatment pond (Table 10). Fish were found to bioconcentrate and dissipate the label more rapidly than the plant material, probably due to the active intake process for the fish, being respiration, compared to the more passive process of absorption for the vegetation.

By day 24, ^{14}C-O_8CDD concentrations in plant material had reached equilibrium in each pond, with corresponding CF values of 380 in the low treatment and 83 in the high treatment pond (Table 10). These values are 3 to 4 orders of magnitude lower than those reported by Corbet et al. (1983), who calculated CF's in the order of 20000 for 1,3,6,8-T_4CDD. Considering that our values were calculated from concentrations of ^{14}C-O_8CDD in floating and rooted vegetation, whereas Corbet's value of 20000 was calculated from 1,3,6,8-T_4CDD concentrations in rooted vegetation only, one can better see the low efficiency of uptake of O_8CDD by the plant material compared to 1,3,6,8-T_4CDD.

Table 10

Concentration of O_8CDD in Fish and Vegetation

Time (days)	Low treatment O_8CDD in water ng/L	CF in Fish	CF in vegetation***	High treatment O_8CDD in water ng/L	CF in Fish	CF in vegetation
0	ND	-	-	ND	-	-
0.01	32.3	78	175	89.2	38	24
0.08	17.4	188	318**	19.3	190	250
0.5	14.1	346	*	24.2	1300	*
1	6.9	490	309	14.1	312	684
2	1.5	857	486	11.9	471	707
4	1.4	1200	635	4.2	450	591
8	1.3	950	6500	4.3	675	545
16	0.5	*	8300	4.1	*	213
24	0.1	*	380	2.2	*	83
34	0.1	*	350	1.9	*	200
51	ND	*	-	0.2	*	-

*** Bioaccumulation in Floating and Rooted vegetation

** Floating vegetation only

* None sampled

ND: None detected

5.0 Sediment Compartment

During the first day post-treatment, ^{14}C-O_8CDD concentrations in the sediment were low in both ponds (Figure 11). These low levels early in the experiment indicated that, following its application on salt crystals, the label had not sedimented directly onto the sediment bed. By day 1, sediments had accumulated relatively small amounts of ^{14}C-O_8CDD and concentrations remained fairly stable in both treatments for up to day 16. By day 16, the surface sediment (top 2 cm) in both treatments started to accumulate the label consistently until day 51, and then for up to 1.5 year post-treatment (day 652). In the high treatment pond, total concentration of ^{14}C-O_8CDD in the sediment was 2.43 ± 0.14 ng/g on day 16 and reached 12.6 ± 0.9 ng/g by day 652. In the low treatment pond, total concentration of the label in the sediment was 1.75 ± 0.08 ng/g on day 16 and reached 8.4 ± 1.1 ng/g by day 652.

Day 16 (September 2) coincided with the beginning of an excessive sedimentation of detritus associated with the end of the season. ^{14}C-O_8CDD associated with decaying plants and/or fish possibly enhanced the concentration of ^{14}C-O_8CDD found in the sediments.

These increases in concentration resulted in a deposition rate (from day 16 up to day 652) of 0.01 ng/g d^{-1} (r^2=0.92) in the low treatment and 0.02 ng/g d^{-1} (r^2=0.97) in the high treatment pond.

As it was observed by Corbet et al. (1983) with the tetra

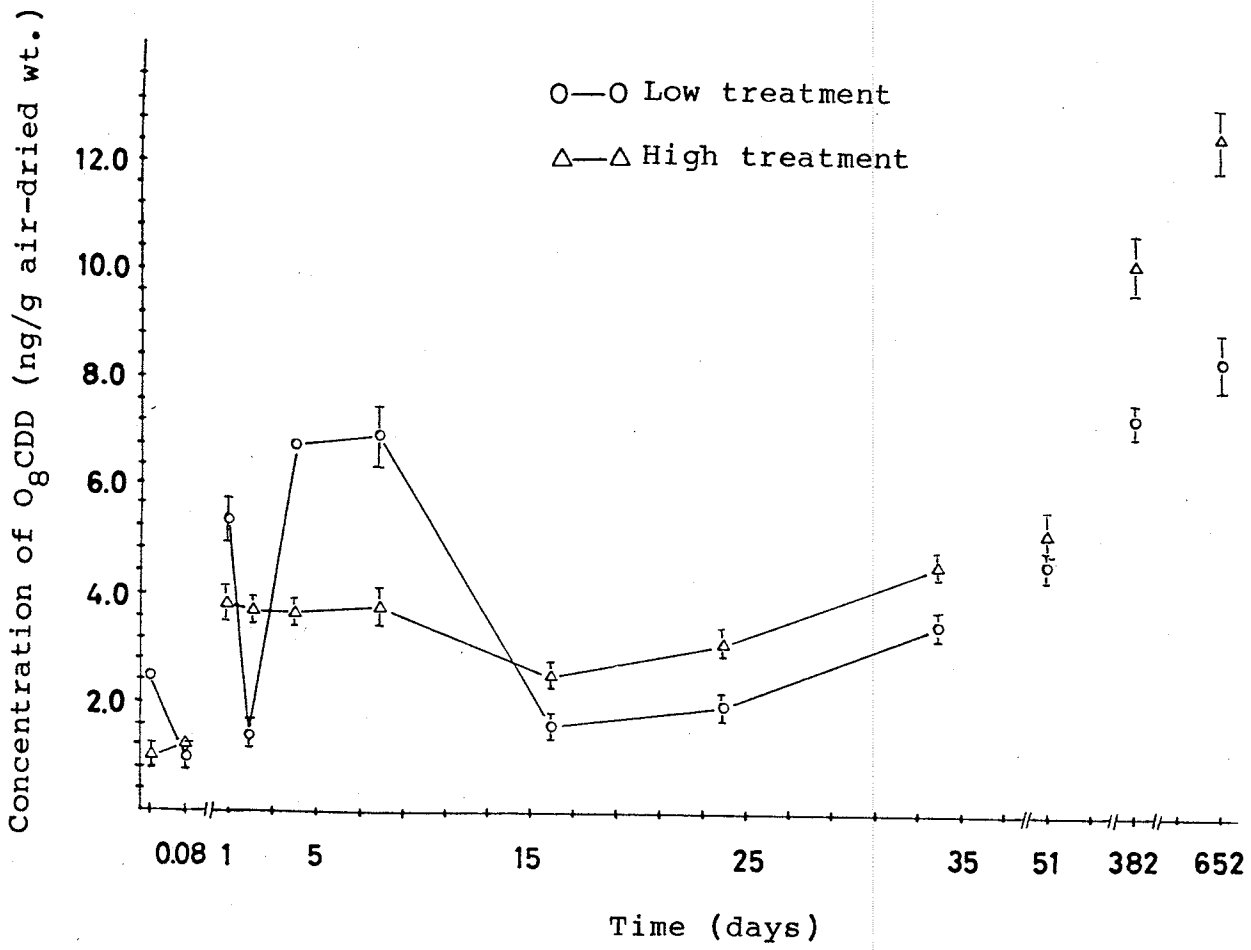


Figure 11. Sediments Levels of O_8CDD

Mean bars represent range

isomer 1,3,6,8-T_4CDD, the pond sediments appear to represent an important storage mechanism of O_8CDD. However, the mechanism appears somewhat more important for O_8CDD, which has been found to accumulate in the sediment for up to 652 days post-treatment.

Recoveries of ^{14}C-O_8CDD from sediment by soxhlet extraction were generally very poor (Table 11). The efficiency of the recovery of organic compounds from sediments may depend on the extraction method, the organic solvent, the type of compound measured and the characteristics of the sediments.

Sporstøl et al. (1983) studied the recovery of a wide range of organic compounds from sediments. Their results suggested that organic compounds found in sediments are generally extracted more effectively by soxhlet and/or ultrasonic extraction. However, the characteristics of the sediments as well as the type of compound being extracted may have been the determining factors responsible for the poor recoveries observed.

Organic particles in aquatic sediments, which originate from decaying organisms, will be porous, non-rigid structures (Bruggeman, 1982). In our study, the organic matter content of the pond sediments approaches 13%. Although it has not been characterized, it can be assumed to be composed of fulvic and humic acids as well as humin fractions.

These various soil fractions may have retained the dioxin tightly enough to make it recalcitrant to extraction. In

Table 11

Accumulation of O_8CDD in sediments

	Time (days)	ng recovered from extraction ng/g air-dried wt. (A)	Total ^{14}C[c] ng/g air-dried wt. (B)	Extraction efficiency (%) (A/B) x 100
Pond 2[a]	0	ND	ND	-
	0.08	1.54	2.50[b]	62
	0.5	0.21 ± 0.01	1.22 ± 0.02	21
	1	3.6 ± 0.1	5.4 ± 0.4	67
	2	0.27 ± 0.01	1.53 ± 0.09	18
	4	3.56	6.78[b]	53
	8	1.2 ± 0.1	6.9 ± 1.0	17
	16	0.32 ± 0.02	1.75 ± 0.08	18
	24	0.61 ± 0.01	2.11 ± 0.06	29
	34	0.875 ± 0.004	3.495 ± 0.054	33
	51	1.21 ± 0.07	4.55 ± 0.08	26
	382	2.7 ± 0.4	7.52 ± 0.45	36
	652	5.2 ± 0.7	8.4 ± 1.1	62
Pond 5[a]	0	ND	ND	-
	0.08	0.16 ± 0.04	1.01 ± 0.06	16
	0.5	0.12	1.08[b]	11
	1	1.2 ± 0.3	4.00 ± 0.33	30
	2	0.76 ± 0.05	3.70 ± 0.06	21
	4	1.01 ± 0.03	3.67 ± 0.04	27
	8	0.51 ± 0.02	3.84 ± 0.32	13
	16	0.83 ± 0.04	2.43 ± 0.14	34
	24	1.05 ± 0.01	3.15 ± 0.11	33
	34	1.37 ± 0.01	4.50 ± 0.05	30
	51	1.5 ± 0.3	5.27 ± 0.32	28
	382	5.1 ± 0.5	10.03 ± 0.56	51
	652	4.3 ± 0.7	12.6 ± 0.9	34

[a] Low treatment (Pond 2) and High treatment (Pond 5).

[b] One duplicate lost, $^{c}14C$ was shown to be ^{14}C-O_8CDD by HPLC analysis.

ND: None detected. Mean values represent range.

other words, the tightly-complexed O_8CDD would have been trapped inside those porous, non-rigid structures, resulting in the poor extraction efficiencies observed.

6.0 Structural Characteristics

As it was discussed in the previous sections, the molecular volume and incumbrance area of O_8CDD may have been responsible for the particular results obtained. This study showed that O_8CDD was poorly concentrated in plants and fish and that the isomer was found to accumulate and be tightly retained in sediments.

The molecular volume of O_8CDD was calculated by summing up contributions from each atom of the molecule (see Table 7). As a result, O_8CDD would have the larger volume of all dioxin isomers, since it has the highest number of chlorine atoms. The molecular volume would then be the same among dioxin congeners since the method used to calculate them is independent of the chlorine substitution pattern. Table 12 presents results which show that the volume of dioxin molecules appears to increase with increasing number of chlorine atoms. The interesting observation that can be made is that O_8CDD appears to have a much larger molecular volume than 1,3,6,8-T_4CDD (T_4CDD congener). Such an observation would be consistent with results obtained by Corbet et al. (1983) and those reported in this thesis, which showed that 1,3,6,8-T_4CDD was more easily concentrated in plants and fish than O_8CDD, possibly because of its

smaller molecular volume.

Table 12

Molecular Volumes of Dioxin Congeners Calculated from Atomic Increments

Congeners	Molecular Volume ($\AA^3$)
O_8CDD	268
H_7CDD	253
H_6CDD	239
P_5CDD	224
T_4CDD	210

The incumbrance area of 1,3,6,8-T_4CDD would also be smaller than of O_8CDD (Figure 12). The incumbrance area of O_8CDD was estimated to be 128 A^2, 103 A^2 for 1,3,6,8-T_4CDD, and 94.8 A^2 for 2,3,7,8-T_4CDD. McKinney and McConnell (1982) reported an incumbrance area of 93 A^2 for 2,3,7,8-T_4CDD. The value of the incumbrance area, as calculated by this method, will differ among congeners (Figure 12) depending on the position of chlorine atoms in the molecule.

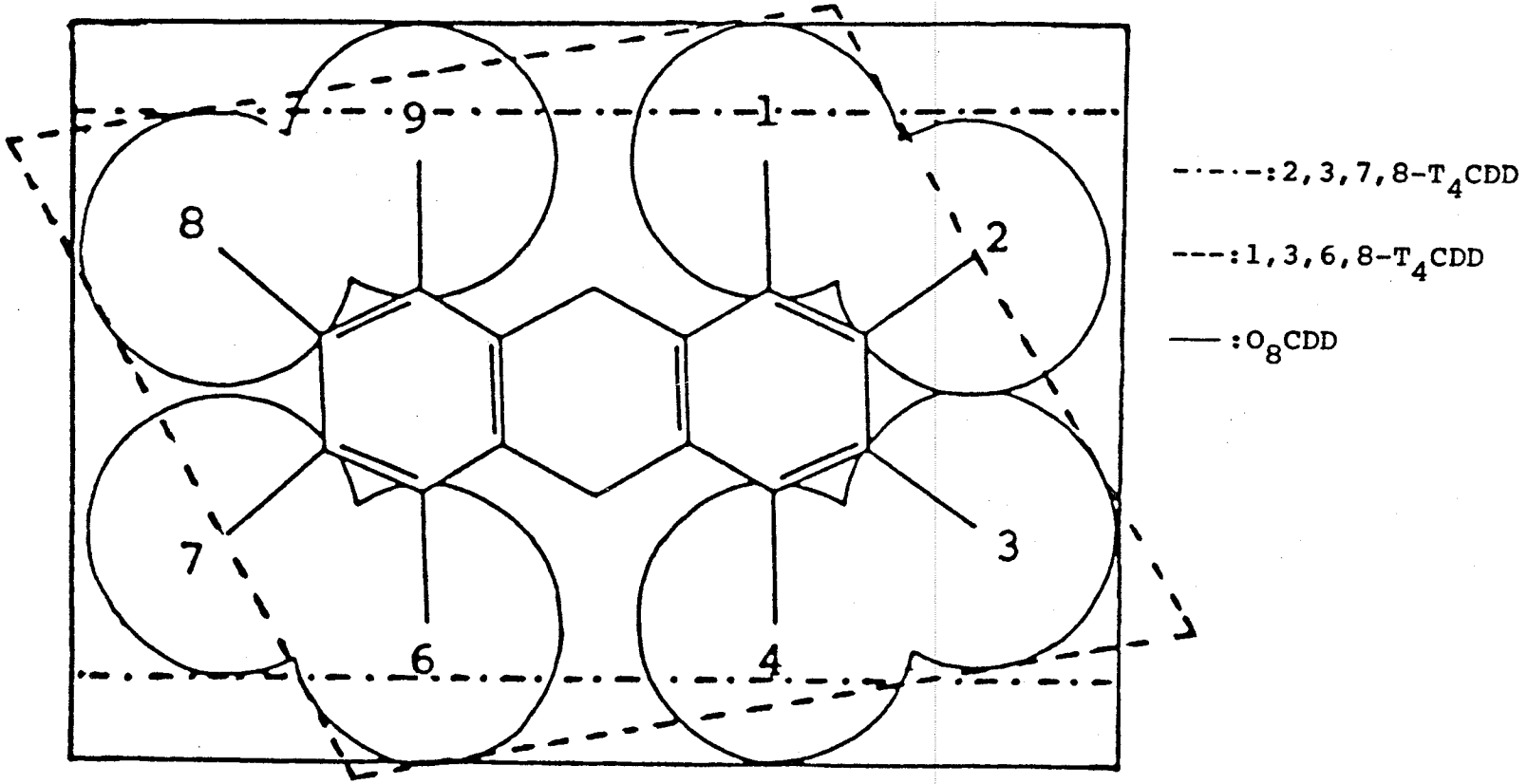


Figure 12. Incumbrance Area of 3 Dioxins

7.0 Mass Balance

Mass balance calculations were attempted to see how much of the applied O_8CDD could be accounted for. Table 13 shows the mass balance at various times post-treatment. The data clearly illustrates that the sediment was a very important sink for O_8CDD in the pond system. After 382 d, most of the remaining ^{14}C was present in bottom sediments, representing 50 to 88% of the amount originally added.

Table 13

Distribution of ^{14}C in Small Ponds Treated with O_8CDD

		Weight (mg) in each compartment*			% in each compartment		
Time	Pond	Water	Sed.	Flora	Water	Sed.	Flora
2h	low	0.08	0.4	0.03	4.7	23.5	1.8
	high	0.09	0.2	0.03	2.6	5.8	0.9
24h	low	0.06	0.9	0.02	3.5	52.9	1.2
	high	0.1	0.7	0.09	2.9	20.6	2.6
16d	low	<0.01	0.3	0.04	<0.6	17.6	2.3
	high	0.02	0.4	<0.01	0.6	11.8	<0.06
24d	low	<0.01	0.4	<0.01	<0.6	23.5	<0.1
	high	0.02	0.5	<0.01	0.6	14.7	<0.3
51d	low	ND	0.8	<0.01	ND	47.1	<0.6
	high	<0.01	0.9	<0.01	<0.3	26.5	<0.3
382d	low	-	1.3	-	-	76.5	-
	high	-	1.7	-	-	50.0	-
652d	low	-	1.5	-	-	88.2	-
	high	-	2.1	-	-	61.8	-

*Quantity in water, sediment and flora calculated by multiplying concentration of total carbon-14 by weight (171700 g of sediment, 26000 g of vegetal matter) or by volume (4.9 m^3 of water) of each compartment.

ND: None detected. (-): Not sampled.

There is a discrepancy in the mass balance in that more is accounted for at later dates. At 2 h posttreatment, 31 % of ^{14}C could be accounted for in the low treatment and only 10 % could be accounted for in the high treatment pond. However, at day 652 posttreatment, 88 % could be accounted for in the low treatment and 62 % in the high treatment pond. It is possible that as the chemical was applied to the water column, it spreaded laterally to the edges of the ponds, where it could have sorbed to sediment and plant material. As a result, very small quantities of O_8CDD could be accounted for at early sampling times. Later in the season, disturbance of the sediment, due to fluctuating water levels as well as freezing and thawing, may have released the O_8CDD in the water column which gradually deposited into the bottom. Combine to this "edge" effect, sedimentation of detrital material, mostly decayed plants, would have occurred during the later sampling times, accounting for the high percentages of O_8CDD recovered at later sampling times.

V CONCLUSIONS

Very small concentrations of O_8CDD in air (pg/L) were detected throughout the experiment but could not be quantitated. Mass balance calculations tend to demonstrate that air was a minor compartment of radioactivity, as 88 % and 62 % of applied O_8CDD could be accounted for on day 652 in the low and high treatment pond respectively.

O_8CDD was found to disappear very rapidly from the water column. The calculated rates of dissipation (0 to 8 d) of 1.75×10^{-2} h^{-1} in the low treatment and 1.55×10^{-2} h^{-1} in the high treatment pond show that dissipation of O_8CDD from the water was rapid at early sampling times. The water compartment accounted for its highest percentages of applied O_8CDD within the first day posttreatment. Fish, which can absorb O_8CDD through their skin, gut and gills, did not appear to be capable of accumulating O_8CDD from the water to a great extent, as calculated CFs were never higher than 1300. Mass balance calculations demonstrated that fish was a minor compartment of radioactivity throughout the study, as less than 1 % of O_8CDD could be accounted for.

The plant material concentrated the label more efficiently but as it was shown with the fish, it was not able to retain the label for long periods of time. Mass balance calculations showed that the plant material accounted for very small percentages of the amount of O_8CDD originally

applied. It is believed that the weight of sediment and volume of water compartments were within 10 to 15 % since dimensions of the ponds were known. The weight of aquatic plants was estimated by multiplying their area by plant density (Wetzel, 1983) and therefore would be subject to considerable error. Another source of error could have been a greater deposition of O_8CDD on plant material and sediment on the sides of the ponds which was not taken into account by sampling the plant material and/or the pond bottom. This has been observed with Fenitrothion (Malis and Muir, 1984) and Deltamethrin (Muir et al., 1985c) in similar outdoor ponds.

O_8CDD persistence was found to be longest in the sediment. O_8CDD was still detected in sediments of each pond at 652 days posttreatment; 8.4 ng/g in the low treatment and 12.6 ng/g in the high treatment pond. The environmental significance of these results may well be that O_8CDD would not be available to fish or either plants but would rather accumulate and persist into sediments of aquatic systems. These pools of accumulated O_8CDD could now be capable of acting as a source for new O_8CDD in the water. Due to long time constants, it will probably take years for the ponds to clean themselves by natural means. Only time will purge the O_8CDD reservoirs in the ponds. The ultimate fate of O_8CDD in aquatic systems under field conditions is in need of further study.

APPENDIX A

ABBREVIATIONS

2,7,-D_2CDD: 2,7-dichlorodibenzo-p-dioxin

DOM: Dissolved organic matter

GC-EC: Gas Chromatography - Electron Capture Detector

H_6CDD: Hexachlorodibenzo-p-dioxin

H_7CDD: Heptachlorodibenzo-p-dioxin

HPLC: High Performance Liquid Chromatography

LSC: Liquid Scintillation Counting

O_8CDD: Octachlorodibenzo-p-dioxin

PCP: Pentachlorophenol

PCDD: Polychlorodibenzo-p-dioxin

P_5CDD: Pentachlorodibenzo-p-dioxin

POM: Particulate Organic Matter

s^{-1}: $second^{-1}$

T_4CP: Tetrachlorophenol

T_4CDD: Tetrachlorodibenzo-p-dioxin

1,3,6,8-T_4CDD: 1,3,6,8-Tetrachlorodibenzo-p-dioxin

2,3,7,8-T_4CDD: 2,3,7,8-Tetrachlorodibenzo-p-dioxin

THF: Tetrahydrofuran

APPENDIX B

Losses in Chromatographic Step

Sample	Pond	Time days	Extracted* ng	Extraction Effici. %	Extracted but lost** ng		Cleanup Effici. %	Tot. ng/g
					Top	Bot.		
Sed.	Low	382	2.7	36	ND	ND	100	7.52
"	High	"	5.1	51	ND	0.8	85	10.03
Fish	Low	1	5.5	79	ND	0.8	85	6.9
"	High	"	6.2	83	ND	1.0	84	7.5
Duck.	Low	0.01	4.1	85	ND	ND	100	4.9
"	High	2	5.8	90	ND	0.5	91	6.5

*ng extracted includes losses from cleanup step

**Losses via chromatographic step (cleanup) checked by combusting top part and bottom (bot.) part of chromatographic column (0.4 to 0.6 g) for remaining ^{14}C.

Duck.: Duckweed (Floating vegetation).

Sed.: Sediment

Tot.: Total concentrations, as reported in results section.

ND: None detected

BIBLIOGRAPHY

L.J. Battan. 1979. Fundamentals of Meteorology. Prentice-Hall, New York, 321 pages.

M.H. Bickel, W.R. Jondorf, S. Muhlebach, and P.A. Wyss. 1983. Adipose Tissue Storage of Polychlorinated Compounds In: Human and Environmental Risks of Chlorinated Dioxins and Related Compounds. R.E. Tucker, A.L. Young, and A.P. Gray, eds. Plenum Press, 477-482.

W.A. Bruggeman. 1982. Hydrophobic Interaction in the Aquatic Environment. In: Handbook of Environmental Chemistry, vol. 2, Part B. O. Hutzinger, ed. Springer-Verlag, New York, 83-102.

W.A. Bruggeman, A. Opperhuizen, A. Wijbenga, and O. Hutzinger. 1984. Bioaccumulation of Super-Lipophilic Chemicals in Fish. Toxicol. Environ. Chem. 7, 173-189.

J.S. Cantrell, N.C. Webb, and A.J. Mabis. 1969. The Identification and Crystal Structure of a Hydropericardium Producing Factor: 1,2,3,7,8,9-hexachlorodibenzo-p-dioxin. Acta Crystallogr., Sect.B. 25, 150-155.

G.G. Choudhry, K. Olie, and O. Hutzinger. 1982. In: Chlorinated Dioxins and Related Compounds. O. Hutzinger, R.W. Frei, E. Merian, and F. Pocchiari, eds. Pergamon Press, 275-301.

R.E. Clement, A.C. Viau, and F.W. Karasek. 1984. Comparison of Solvent Extraction Efficiencies for Toxic Organic Compounds on Flyash. Can. J. Chem. 62, 2629-2633.

B.L. Corbet, G.R.B. Webster, and D.C.G. Muir. 1983. Fate of 1,3,6,8-TCDD in an Outdoor Aquatic System. Chemosphere, 12, 523-527.

N.O. Crossland, and C.J.M. Wolff. 1985. Fate and Biological Effects of Pentachlorophenol in Outdoor Ponds. Environ. Toxicol. Chem. 4, 73-86.

M.R. Cull, A.J. Dobbs, and N. Williams. 1983. Polychlorodibenzo-p-dioxin (PCDDs) in Commercial Pentachlorophenol used in Wood Preservation. Chemosphere, 12, 483-485.

M.R. Cull, and A.J. Dobbs. 1984. Long Term Changes in Polychlorodibenzo-p-dioxin Concentrations in Wood Treated with Technical Pentachlorophenol. Chemosphere, 13, 1091-1099.

M.R. Cull, A.J. Dobbs, M. Goudot, and N. Schultz. 1984. Polychlorodibenzo-p-dioxins and Dibenzofurans in Technical Pentachlorophenol - Results of a Collaborative Analytical Exercise. Chemosphere, 13, 1157-1165.

J.M. Czuczwa, and R.A. Hites. 1984. Environmental Fate of Combustion - Generated Polychlorinated Dioxins and Furans Environ. Sci. Technol. 18, 444-450.

A.J. Dobbs, and C. Grant. 1981. Octachlorodibenzo-p-dioxin in Wood Treatment Materials and Treated Wood. Chemosphere 10, 1185-1193.

A.J. Dobbs, and M.R. Cull. 1982. Volatilisation of Chemicals Relative Loss Rates and the Estimation of Vapour Pressure Environ. Pollut. Ser. B. 3, 289-298.

A.J. Dobbs, and N. Williams. 1983. Fat Solubility - A Property of Environmental Relevance? Chemosphere, 12, 97-104.

Dow Chemical, U.S.A., 1978. The Trace Chemistries of Fire - A Source of and Routes for the Entry of Chlorinated Dioxins into the Environment. 46 pages.

J.T. Edward. 1970. Molecular Volumes and the Stokes Einstein Equation. J. Chem. Education, 47, 261-270.

M.P. Esposito, T.O. Tiernan, and F.E.Dryden. 1980. Dioxins. United States Environmental Protection Agency. 251 p.

J.A. Goldstein, M. Friesen, R.E. Linder, P. Hickman, J.R. Hass, and H. Bergman. 1977. Effects of Pentachlorophenol on Hepatic Drug-Metabolizing Enzymes and Porphyria Related to Contamination with Chlorinated Dibenzo-p-dioxins and Dibenzofurans. Biochem. Pharmac. 26, 1549-1557.

J.L. Hamelink, and R.C. Waybrant. 1976. DDE and Lindane in a Large-Scale Model Lentic Ecosystem. Trans. Amer. Fish. Soc. 105, 124-134.

C.W. Heckman. 1982. Pesticide Effects on Aquatic Habitats. Environ. Sci. Technol. 16, 48A-57A.

A.R. Isensee, and G.E. Jones. 1975. Distribution of 2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD) in Aquatic Model Ecosystem. Environ. Sci. Technol. 9, 668-672.

P.A. Jones. 1981. Les Chlorophénols et leurs Impuretés dans l'Environnement Canadien. Service de la Protection de l'Environnement, Rapport SPE-EC-81-2F, 321 pages.

F. Korte. 1983. Ecologic Chemistry of Dioxins and Related Compounds. In: Accidental Exposure to Dioxins; Human Health Aspects. F. Coulston, and F. Pocchiari, eds. Academic Press, New York, 171-181.

L.L. Lamparski, R.H. Stehl, and R.L. Johnson. 1980. Photolysis of Pentachlorophenol-Treated Wood. Chlorinated Dibenzo-p-dioxin Formation. Environ. Sci. Technol. 14, 196-200.

P.F. Landrum, S.R. Nihart, B.J. Eadie, and W.S. Gardner. 1984. Reverse - Phase Separation Method for Determining Pollutant Binding to Aldrich Humic Acid and Dissolved Organic Carbon of Natural Waters. Environ. Sci. Technol. 18, 187-192.

P.S. Liss, and P.G. Slater. 1974. Flux of Gases Across the Air - Sea Interface. Nature, 247, 181-184.

J.W.A. Lustenhouwer, K. Olie, and O. Hutzinger. 1980. Chlorinated Dibenzo-p-dioxins and Related Compounds in Incinerators Effluents: A Review of Measurements and Mechanisms of Formation. Chemosphere, 9, 501-522.

F.K. Lutgens, and E.J. Tarbuck. 1979. The Atmosphere: An Introduction to Meteorology. Prentice-Hall, pp.413.

J.W.Lyman. 1982. Octanol/Water Partition Coefficient. In: Handbook of Chemical Property Estimation Methods. J.W. Lyman, W.F. Reehl, and D.H. Rosenblatt, eds. McGraw-Hill, New York, 1-1 to 1-54.

D. Mackay, W.Y. Shiu, and R.P. Sutherland. 1979. Determination of Air - Water Henry's Law Constants for Hydrophobic Pollutants. Environ. Sci. Technol. 13, 333-337.

D. Mackay. 1980. Solubility, Partition Coefficients, Volatility, and Evaporation Rates. In: Handbook of Environmental Chemistry, vol.2, Part A. O. Hutzinger, ed. Springer-Verlag, 31-45.

D.J. Madder. 1978. The Disappearance from, Efficacy in and Effect on Non - Target Organisms of Diflubenzuron, Methoprene, and Chlorpyrifos in a Lentic Ecosystem. M. Sc Thesis, University of Manitoba, Winnipeg, Manitoba.

G.P. Malis and D.C.G. Muir. 1984. Fate of Fenitrothion in Shaded and Unshaded Ponds. ACS Symposium Series No. 238 Chemical and Biological Controls in Forestry, W.Y. Garner and J. Harvey Jr., Eds.

J. McKinney, and E. McConnell. 1982. Structural Specificity and Dioxin Receptor. In: Chlorinated Dioxins and Related Compounds. O. Hutzinger, R.W. Frei, E. Merian, and F. Pocchiari, eds. Pergamon Press, New York, 367-381.

D.C.G. Muir, K. Marshall, and G.R.B. Webster. 1985. Bioconcentration of PCDD's by Fish: Effects of Molecular Structure and Water Chemistry. Chemosphere 14, 829-833.

D.C.G. Muir, A.L. Yarechewski, and G.R.B. Webster. 1985a. Bioconcentration of Four Chlorinated Dioxins by Rainbow Trout and Fathead Minnows. In: Proceedings of the 8th ASTM Symposium on Aquatic Toxicology. In Press.

D.C.G. Muir, A.L. Yarechewski, A. Knoll, and G.R.B. Webster. 1985b. Bioconcentration and Disposition of 1,3,6,8-T_4CDD and O_8CDD by Rainbow Trout and Fathead Minnows. Environmental Toxicology and Chemistry. In Press.

D.C.G. Muir, G.P. Rawn, and N.P. Grift. 1985c. Fate of the Pyrethroid Insecticide Deltamethrin in Small Ponds: A Mass Balance Study. J. Agric. Food Chem., 33, 603-609.

C.A. Nilsson, A. Norstrom, K. Anderson, and C. Rappe. 1978. Impurities in Commercial Products Related to Pentachlorophenol. In: Pentachlorophenol; Chemistry, Pharmacology, and Environmental Toxicology. K.R. Rao, ed. Plenum Press, New York, 313-324.

D.H. Norback, J.F. Engblom, and J.R. Allen. 1975. Tissue Distribution and Excretion of Octachlorodibenzo-p-dioxin in the Rat. Toxicol. App. Pharmacol. 32, 330-338.

NRCC. 1981. Polychlorinated Dibenzo-p-dioxins: Criteria for their Effects on Man and his Environment. Report no 18574 251 pages.

A. Poland. E. Glover, and A.S. Kende. 1976. Stereospecific, high Affinity Binding of 2,3,7,8-Tetrachlorodibenzo-p-dioxin by hepatic cytosol. Evidence that the Binding Species is Receptor for Induction of Aryl Hydrocarbon Hydroxylase. J. Biol. Chem. 251, 4936-4946.

A. Poland, W.F. Greenlee, and A.S. Kende. 1979. Studies on the Mechanism of Toxicity of the Chlorinated Dibenzo-p-dioxins and Related Compounds. Ann. NY Acad. Sci. 320, 214-230.

K.R. Rao, ed. 1978. Pentachlorophenol: Chemistry, Pharmacology, and Environmental Toxicology. Plenum Press, 402 p.

S.Safe, L. Robertson, T. Sawyer, A. Parkinson, S. Bandiera L. Safe, and M. Campbell. 1983. PCDDs and Related Compounds: Metabolism and Biochemistry. In: Human and Environmental Risks of Chlorinated Dioxins and Related Compounds. R.E.Tucker, A.L. Young, and A.P. Gray, eds. Plenum Press, New York, 393-403.

L.P. Sarna, P.E. Hodge, and G.R.B. Webster. 1984. Octanol-Water Partition Coefficients of Chlorinated Dioxins and Dibenzofurans by Reverse - Phase HPLC using several C18 Columns. Chemosphere, 13, 975-983.

B.A. Schwetz, J.M. Norris, G.L. Sparschu, V.K. Rowe, P.J. Gehring, J.L. Emerson, and C.G. Gerbig. 1973. Toxicology of Chlorinated Dibenzo-p-dioxins. Environ. Health Perspect. 9, 87-99.

J. Singh. 1981. Unpublished Results. Laboratory Services Division, Agriculture Canada.

J. Solomon, and D.C.G. Muir. 1981. Use of Micro - Analytical Technique for the Determination of Chlorinated Hydrocarbons in Fish Samples. Presented at the AOAC Workshop, May 12-14, Ottawa, Canada.

A. Spacie, and J.L. Hamelink. 1982. Alternative Models for Describing the Bioconcentration of Organics in Fish. Environ. Toxicol. Chem. 1, 309-320.

P. Spangler. 1983. Unpublished Results. Pathfinder Laboratory. Inc St-Louis, Mo.

S. Sporstøl, N. Gjos, and G. Carlberg. 1983. Extraction Efficiencies for Organic Compounds found in Sediments. Anal. Chim. Acta, 151, 231-235.

D.L. Stalling, L.M. Smith, J.D. Petty, J.L. Hogan, C. Rappe, and H.R. Buser. 1983. Residues of Polychlorinated Dibenzo-p-Dioxins and Dibenzofurans in Laurentian Great Lakes Fish. In: Human and Environmental Risks of Chlorinated Dioxins and Related Compounds. R.E. Tucker, A.L. Young, and A.P. Gray, eds. Plenum Press, New York, 221-240.

K. Sugiura, M. Aoki, S. Kaneko, I. Daisaku, Y. Komatsu, H. Shibuya, H. Suzuki, and M. Goto. 1984. Fate of 2,4,6-Trichlorophenol, Pentachlorophenol, p-Chlorobiphenyl, and Hexachlorobenzene in an Outdoor Experimental Pond: Comparison between Observations and Predictions based on Laboratory Data. Arch. Environ. Contam. Toxicol. 13, 745-758.

L.J. Thibodeaux. 1979. Chemodynamics: Environmental Movement of Chemicals in Air, Water, and Soil. John Wiley & Sons, New York, 501 pages.

G. Tsushimoto, F. Matsumura, and R. Sago. 1982. Fate of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) in an Outdoor Pond and in Model Aquatic Ecosystems. Environ. Toxicol. Chem. 1, 61-68.

M.Th.M. Tulp, and O. Hutzinger. 1978. Some Thoughts on Aqueous Solubilities and Partition Coefficients of PCB, and the Mathematical Correlation between Bioaccumulation and Physico - Chemical Properties. Chemosphere, 7, 849-860.

M.Th.M. Tulp, and O. Hutzinger. 1978a. Rat Metabolism of Polychlorinated Dibenzo-p-dioxins. Chemosphere, 9, 761-768.

US Environmental Protection Agency, Science Advisory Board. 1978. Estimated Vapour Pressure Included in Draft Report of Ad Hoc Study Group on Pentachlorophenol Contaminants.

G.D. Veith, D.L. Defoe, and B.V. Bergstedt. 1979. Measuring and Estimating the Bioconcentration Factor of Chemicals in Fish. J. Fish. Res. Board Can. 36, 1040-1048.

C.T. Ward, and F. Matsumura. 1978. Fate of 2,3,7,8-TCDD in a Model Aquatic Environment. Arch. Environm. Contam. Toxicol. 7, 349-357.

J.S. Wassom, J.E. Huff, and N. Loprieno. 1977-1978. A Review of the Genetic Toxicology of Chlorinated Dibenzo-p-dioxins. Mutation Res. 47, 141-160.

G.R.B. Webster et al. 1981. Unpublished Results.

G.R.B. Webster, L.P. Sarna, and D.C.G. Muir. 1983. Kow of 1,3,6,8-TCDD and OCDD by Reverse - Phase HPLC. ACS Meeting on Chlorodioxins and Chlorofurans. Washington, D.C. August 1983.

G.R.B. Webster. 1984. Availability and Bioconcentration of Polychlorinated Dibenzo-p-dioxins Associated with Sediment Organic Matter. Final Report, Year 2, OSU 82-00277, DSS. 44 pages.

R.G. Wetzel.1983. Limnology. Saunders College Publishing, New York, 767 pages.

D.T. Williams, H.M. Cunningham, and B.J. Blanchfield. 1972. Distribution and Excretion Studies of OCDD in the Rat. Bull. Environ. Contam. Toxicol. 7, 57-62.

R.S. Yockim, A.R. Isensee, and G.E. Jones. 1978. Distribution and Toxicity of TCDD and 2,4,5-T in an Aquatic Ecosystem. Chemosphere, 3, 215-220.