AG11

NRA WESSEX REGION
LIBRARY

Interim Report R & D Project 240
Bioaccumulation as an Approach for Detection and Monitoring of Red List Trace Organic Contaminants: A Literature Review

ERL (North)
October 1992
R & D/240/11/Y

CONSERVATION

Research Contractor:
ERL (North)
No 1 Industrial Estate
Medomsley Road
Consett
Co.Durham
DH8 6SS

National Rivers Authority
Rivers House
Waterside Drive
Almondsbury
Bristol
BS12 4UD

ENVIRONMENT AGENCY

NATIONAL LIBRARY &
INFORMATION SERVICE

~~SOUTH WEST REGION~~
HEAD OFFICE
~~Manley House, Kestrel Way~~
~~Exeter EX2 7LQ~~
BRISTOL

ENVIRONMENT AGENCY
081523

National Rivers Authority
Rivers House
Waterside Drive
Almondsbury
Bristol
BS12 4UD

Tel: 0454-624400
Fax: 0454-624409

(c) National Rivers Authority 1992

All rights reserved. No part of this publication may be reproduced, stored in a retrieval system, or transmitted, in any form or by any means, electronic, mechanical, photocopying, recording or otherwise without the prior permission of the National Rivers Authority.

Dissemination Status
Internal : Released to Regions
External : Released to Public Domain

Research Contractor

This document was produced under R & D Project 240 by:

ERL (North)
No 1 Industrial Estate
Medomsley Road
Consett
Co.Durham
DH8 6SS

Tel: 0207-507254
Fax: 0207-580026

NRA Project Leader
The NRA's Project Leader for R & D Project 240 :
Colin Urquhart - Yorkshire Region

Additional Copies
Further copies of this document may be obtained from Regional R & D Co-ordinators or the R & D Section of NRA Head Office.

Document Status and Use Note

This note provides details of how the attached document from the R&D programme is to be used and serves as a record of its status. Consequently, this note should not be removed from this document.

1. Document

Title: Bioaccumulation as an approach for detection and monitoring of Red List Organic Contaminants : A Literature Review.

Distribution Instructions

One copy to poject contacts in the Regions (see attached circulation list).

Dissemination Status	Internal	Released to Region	External	Public Domain

2. Document Status and Intended Use

Details (should refer to where the document stands in relation to other documents and how it is to be used)

This document is for Information Only.

3. Approval For Status and Use

Project Leader	C. Urquhart	Topic Leader	–
Commissioner	–	Group Chairman[1]	–

Notes: [1] Signature of Chairman of Working Group if appropriate.

Date Output Sent: 9/2/93

This document may have been distributed prior to the end of research stage assessment and before any decision has been made concerning its implementation.

Approval for permanent project outputs will be required from Project Leader/ Topic Leader/ Commissioner/Group Chairman. Less signatures will be required for draft or interim outputs.

CONTENTS

1 INTRODUCTION

1.1 General Introduction to Biomonitoring

Throughout the UK and Europe the number of reported water pollution incidents involving trace organic contaminants is increasing. This together with tightening legislative controls on water quality and improvements in the technology that assists in the detection and measurement of trace organic pollutants in water means that the need for monitoring these substances is essential.

There are fundamentally four approaches to detecting and monitoring trace organic pollutants in water. These are either:

directly through the:

- sampling and direct measurement of concentrations in water;

or indirectly through the:

- sampling and analysis of concentrations in sediments;
- measurements of concentrations taken up by synthetic materials;
- use of biomonitoring.

Biomonitoring or the use of biological indicator organisms can be further divided into three different approaches either:

- using organisms as indicators of contamination through either their presence or absence as a direct response to the pollutant;
- using organisms in bioassays and measuring their biochemical response to pollutants;
- using the measurement of tissue concentrations of pollutant.

Most water quality monitoring programmes involve one or more of these various approaches and the majority of water management bodies accept that any monitoring programme or system will comprise a blend of biological with other procedures.

This review will concentrate on an evaluation of bioaccumulation as an approach to detecting and monitoring trace organic pollutants.

1.2 Interest of the National Rivers Authority (NRA)

In the UK the NRA has become increasingly concerned about contamination of surface and groundwaters by organic pollutants, in particular those featured in the priority Red List as issued by the Department of the Environment (see 2.1 below). Major problems associated with trace organics have become evident in the UK with, in the last decade the emphasis switching from the organochlorine compounds which were notorious in the past (Cope, 1966; Mellanby 1970; and Hellawell, 1986) to other pesticides such as herbicides (Croll, 1991) and organophosphorus insecticides (Hellawell, 1986) as well as tributyl tin compounds (Langston et al, 1990; Waldock et al, 1983, 1987).

Sources of these trace organic contaminants can often be both diffuse (ie non point-source) and intermittent and the concentrations present in the water have often been at or below the limits of detection for the more routinely used methods and instrumentation.

The attraction in the use of tissue concentration measurement is discussed in more detail in Section 4. Essentially the accumulation and subsequent bioconcentration of trace organic substances to levels which are higher and easier to measure and the integration of episodic spills or discharges enables substances to be detected after the discharge has ceased. These features of the approach have an additional attraction in that they have a potential for cost savings over the more traditional water analysis programmes .

Bioaccumulation techniques for the detection and monitoring of trace organics are already in use within the water industry. In France, for example, Mouvet and co-workers have developed river monitoring techniques for the river authorities of the Seine-Nomandie and Rhine-Meuse basins (Mouvet et al, 1985, 1986). The techniques are also currently being used by the NRA (see Annex 1) but variation in the methods of sampling and analysis, interpretation of results and application have hindered the effective use and comparison of data.

If the approach is to be adopted successfully and used widely by the NRA as a practical water quality management tool there is a need to:

- standardise methods into practical and usable codes of practice, and
- understand more fully the applicability, interpretation and significance of the data obtained.

1.3 Objectives of the Review

With reference to the points made above, this review aims to assess the current literature on the use of bioaccumulation as an approach to monitoring Red List organic contaminants in both fresh and estuarine waters.

A number of key questions will be addressed within the review, in particular the following:

- What are the advantages of the approach for detecting trace organic pollutants?
- How applicable is the approach for monitoring trends in the concentrations of trace organic pollutants?
- What are the levels of confidence that can be specified in the results of tissue analysis?
- How can the data be best interpreted and what does it actually tell you?

2 CHARACTERISTICS OF RED LIST TRACE ORGANICS WITH REFERENCE TO BIOACCUMULATION POTENTIAL

2.1 Introduction

The organic contaminants that this report is most concerned with are those that are included in the Government's Red List.

The main groups of compounds are:

- Organochlorine Pesticides
- Polychlorinated Biphenyls (PCBs)
- Pentachlorophenols
- Organophosphorus Pesticides
- Herbicides
- Organic Solvents
- Triorganotin Compounds.

Inclusion on the Red List means that they are identified as 'substances which represent the greatest threat to the aquatic environment because of their toxicity, persistence and capacity for bioaccumulation' (DoE, 1988). Table 2.1a shows the Red List organic compounds. There is a considerable number of literature reviews relating to representatives of the Red List trace organics and the reader is directed particularly to the following with reference to bioaccumulation:

- Organochlorines and PCBs
  - for the period up to 1980, see the reviews of Phillips (1978 and 1980);
  - for the period to 1986 covering these compounds in freshwaters, see the review of Hellawell (1986);
  - for reviews including ecotoxicology and bioaccumulation up to 1989, see reviews in World Health Organisation series (WHO, 1989 a and b).
- Organophosphorus compounds
  - for a review including toxicology up to 1986 see WHO (1986).

- Herbicides
  - for a review of literature to 1987 including surface water chemistry see the reviews of Hellawell (1986) and Bowmer (1987).

Table 2.1a The Red List Trace Organic Compounds

Organochlorine Pesticides
Gamma-hexachlorocyclohexane (Lindane)
DDT
Pentachlorophenol (PCP)
Aldrin
Dieldrin
Endrin
Endosulfan
Organophosphorus Pesticides
Dichlorvos
Azinphos-methyl
Fenitrothion
Malathion
Polychlorinated Biphenyls (PCBs)
Herbicides
Trifluralin
Atrazine
Simazine
Organic Solvents
Hexachlorobutadiene (HCBD)
1,2-Dichloroethane
Trichlorobenzene
Triorganotin Compounds
Tributyltin Oxide
Triphenyltin Acetate
Triphenyltin Chloride
Triphenyltin Hydroxide

Table 2.1b *Possible Sources of Red List Trace Organics to the Aquatic Environment*

Type of Pollution	Type of Compound(s)	Source
point source		
(i) Direct discharge, spillage or via sewage treatment works.	Insecticides / herbicides / fungicides	Agro-chemical manufacturers.
	Insecticides	Textile manufacturers.
	Insecticides	Sheep dip waste waters.
	Wood preservatives	Timber treatment plants.
	Organic solvents / triorganotins	Chemical manufacturers.
(ii) Leakage / leachate from waste disposal sites.	Insecticides / PCBs	Migration of contaminants from hazardous waste disposal sites via groundwaters or leachate entering rivers.
(iii) Disposal of sewage sludge to estuaries or sea.	Insecticides / PCBs	Accumulated in sludge from industrial waste water.
(iv) Illegal dumping of wastes.	All	Migration from waste tips in ground waters, leachate or direct discharge.
(v) Leakage / leachate.	PCBs	Scrapyards where transformers containing dielectric fluids have been broken up.
(vi) Migration from contaminated land sites	all	Migration from operational or derelict sites. High risks during reclamation of derelict land.
non-point source		
	Insecticides / herbicides / fungicides	Agriculture, forestry (run-off; volatilization).
	Wood preservatives / insecticides	Timber treatment, domestic wastes.
	PCBs and related degradation products	Aerosol distribution from poorly operated incinerators.
	Volatile organochlorine compounds	Atmospheric deposition.
	Triorganotin compounds	Leaching from antifouling paints.

Table 2.1b summarises the possible sources of Red List organics in relation to the type of pollution involved, type of compound and its general source.

Table 2.1c summarises the general characteristics of organic chemicals that exhibit substantial bioaccumulation potential.

Table 2.1c General Characteristics of Organic Chemicals that Exhibit Substantial Bioaccumulation (after Connell 1988)

Characteristic	Features giving Bioaccumulation
Chemical Structure	High proportion of C-C(Aliphatic), C-C(Aromatic), C-H, and C-Cl Bonds
Molecular Weight	<Approx 300 with a decline in capacity>300
Stability	Resistant to degradation
Log K_{ow}	2-5 to 6 with a declining capacity >300
Water Solubility	18-0.02 to 0.002 mol M^{-3} with declining capacity at lower values
Degree of Ionization	Very low

2.2 Properties in relation to transport and mobility

The formulation, solubility and chemical and physical properties that determine the persistence of herbicides, insecticides, or similar hydrocarbons, play a major role in determining losses from agricultural land and contaminated soils due to run-off, and consequent transport and mobility within the aquatic habitat. Pesticide persistence is usually measured in terms of half-life ($t_{½}$), the time required for 50% of the pesticide to disappear.

A classification of pesticide persistence in soils based on the rate of disappearance of the solvent-extractable parent compound under aerobic laboratory incubation conditions has been developed by Rao and Davidson (1980). A summary of this is given in Table 2.2a.

Table 2.2a ***Classification of Pesticide Persistence in Soils Based on Rate of Disappearance of the Solvent-Extractable Parent Compound (after Rao and Davidson, 1980).***

Non-Persistent ($t_{1/2}$ < 20 days)	**Moderately Persistent (20 days > $t_{1/2}$ < 100 days)**		**Persistent ($t_{1/2}$ > 100 days)**
2, 4-D	Atrazine	Phorate	Trifluralin
2, 4, 5-T	Simazine	Carbofuran	Bromacil
Dicamba	Terbacil	Carbaryl	Picloram
Dalapon	Linuron	Aldrin	Paraquat
Methyl parathion	TCA	Dieldrin	DDT
Malathion	Glyphosate	Endrin	Chlordane
Captan	Parathion	Heptachlor	Lindane (gamma-HCH)
	Diazinon	PCP	
	Fonophos		

Basic pesticides are immobile at low pH, (pH 6 - 8) but can be mobile at high pH depending on water solubility and chemical structure. Acidic pesticides are mobile at normal soil pH, but arsenate and phosphate pesticides are usually immobile due to their tendency for soil absorption. The non-ionic pesticides are all usually absorbed to some extent in soil, but the degree of absorption depends on water solubility, vapour pressure and chemical structure.

2.3 Organochlorine Compounds

Organochlorine insecticides were the first synthetic organic pesticides to be developed, with DDT and lindane being probably the best known. Their persistence in the environment was at first considered an advantage following their introduction in the 1940s, but later in the 1960s it became evident that they were being detected generally within the environment and often well away from application sites. The water solubility and persistence of the main organochlorine compounds is given in Table 2.3a.

The low solubility of these compounds in water but high solubility in lipids is an important factor in their bioconcentration since they will partition preferentially in tissue lipids thus facilitating their retention.

Table 2.3a ***Water Solubility and Persistence of Organochlorine Insecticides in Soil (after Edwards, 1973)***

Compound	Approximate water solubility (mgl^{-1})	Approximate half-life (years)	Approximate mean time for 95% disappearance of residues / year
DDT	0.001 - 0.04	2.8	10
Dieldrin	0.1 - 0.25	2.5	8
Endrin	0.1 - 0.25	2.2	7
Lindane	7.0 - 10.0	1.2	6.5
Chlordane	very low	1.0	4
Heptachlor	very low	0.8	3.5
Aldrin	0.01 - 0.2	0.3	3

Biomagnification is usually an absorption process that eventually leads to an equilibrium distribution of the pesticide between the organisms' tissues and water. This equilibrium is achieved more rapidly by direct uptake than through food chains.

2.4 Polychlorinated Biphenyls

PCBs are highly lipophilic, and hence of significance to biota, with similar ecosystem cycling and potential pollution problems as the somewhat chemically similar organochlorine insecticide DDT. They have extremely low water solubility. The high lipophilic property of PCBs contributes to their pronounced bioaccumulation. In a comprehensive literature review of the process of uptake and accumulation of PCBs in the aquatic environment, Falkner and Simonis (1982) concluded that, apart from higher vertebrates, bioconcentration in food chains was far less important than direct uptake from water.

The main factors which contribute to their persistence in the environment and hence their bioaccumulation potential are:

- pronounced non-ionic and lipophilic nature;
- low water solubility;
- low volatility;
- resistance to degradation;
- hydrophobic character;

- readiness to adsorb onto particles or sediments.

2.5 Pentachlorophenol (PCP)

Although the ionised form of PCP has a relatively high volatility and water solubility, it is readily adsorbed by soils and sediments and is also bioaccumulated (WHO, 1987). It is relatively stable in the environment and is highly toxic to nearly all forms of life as an inhibitor of oxidative phosphorylation (Lu et al 1978). Exposure can lead to uptake by aquatic organisms, and the problem is compounded by the persistence of PCP and PCP-impurities and degradation products, many of which are also highly toxic (Pierce and Victor, 1978). Bioaccumulation takes place from the surrounding water due to the solubility of PCP but biomagnification along the food chain also contributes to the overall levels accumulated (Lu et al 1978). PCP has been shown to persist in water and fish for over six months following a spill of wood-treatment wastes and for over two years in leaf litter and sediment (Pierce and Victor, 1978).

2.6 Organophosphorus Compounds

It seems that through degradation most organophosphorus pesticides are rapidly lost from food crops and are usually barely detectable four weeks after application, although the exact rate of loss depends on the weather conditions (WHO 1986). Organophosphorus insecticides are therefore not very stable in aqueous media and all are subject to degradation by hydrolysis and oxidation yielding water-soluble products (mainly mono- or di-substituted phosphoric or phosphoric acids or their thio analogues). The half-life at neutral pH may vary from a few hours for dichlorvos to several weeks for parathion. Certain constituents of soil and river water may catalyse degradation. There is also evidence that light, suspended particulates and bacteria all contribute to the degradation (WHO, 1986).

The early work on the potential for bioaccumulation of organophosphate insecticides, (eg. Chisholm et al, 1955; Lichtenstein and Schulz; 1964) indicated that this group of compounds was generally short-lived in the environment and did not bioaccumulate to any serious extent. However, Cowart et al (1971) working on the rate of hydrolysis of seven organophosphate pesticides indicated that although chemical alterations of such compounds through oxidation or hydrolysis may be rapid (measured in hours or days), the decomposition products may not be harmless. Faust and Gomma (1972) reviewed the mechanisms of toxicity and persistence of organophosphate insecticides and concluded that under various natural ranges of environmental conditions in aquatic systems, these compounds can be quite persistent with a half-life of approximately 108 days for parathion.
While malathion is a widely used organophosphate insecticide entering surface waters in various ways (Cook et al, 1976), residue interpretation is difficult due to the toxicity of the "persistent" metabolite (malaoxan) which is not easily identified in tissues. In terms of higher organism accumulation it is unlikely that biomagnification occurs since compounds are unlikely to survive the hydrolytic process of consumption and digestion to be stored

3 MECHANISMS OF BIOACCUMULATION

3.1 Introduction

Aquatic species differ from terrestrial species, in that pollutants may be taken up from, and "excreted" into, the ambient water. Consequently, absorption of pollutants can occur via a number of pathways, namely from solution, suspended particles, sediments and food.

A variety of terms have been used to describe the pathwyas or processes by which aquatic organisms take up trace organic compounds. Moriarty (1988) divides these processes into three basic types:

Bioconcentration: the increase of pollutant concentration from water when passing directly into aquatic species;

Bioaccumulation: this has a similar meaning to bioconcentration but indicates the combined intake from food as well as water;

Biomagnification: the increase in concentration of pollutant in animal tissues in successive members of a food chain.

These three phenomena are discussed in the following sections under the headings of accumulation from water, accumulation from sediments and predator prey relationships.

3.2 Accumulation from Water (Bioconcentration)

A good indication of an organism's ability to bioaccumulate organic contaminants may be given by the octanol-water partition coefficient K_{ow} (Freed and Chiou, 1982). This is based on the fact that octanol has the same hydrophobic properties as the lipids in cells upon which dermal absorption is dependent. The coefficients are therefore essentially an indication of the lipophilicity of the compound. Ernst (1977) showed that the rates of uptake of chlorinated pesticides by *Mytilus edulis* (common mussel) decreased in the order DDD > endrin > heptachlor epoxide > dieldrin and endosulfan > alpha-HCH > gamma-HCH. Decreasing rate was closely related to increasing water solubility, presumably because there is a significant correlation between low water solubility and high lipid-water partitioning (Metcalf 1977).

Walker (1975) considers that rapid surface adsorption or partitioning of the lipoprotein cell membrane probably provides the pathway for diffusion of inorganic pollutants into the organism and molecules with a high liposolubility are usually the most readily absorbed.

At equilibrium bioconcentration, the direct transfer of lipophilic compounds from water to biota can be characterised by the BCF (Walker 1987) where:

$$\text{BCF} = \frac{\text{concentration in whole organism}}{\text{concentration in ambient medium}}$$

Several experimental studies (eg. Neely et al, 1974; Ernst, 1977; Geyer et al, 1982) with ranges of lipophilic compounds have shown either:

- good correlation between BCFs and K_{ow} or;
- good inverse correlations between BCFs and values for water solubility.

Bioconcentration factors (BCF) for PCBs of 50,000 or more are widely reported for fish (Mayer et al, 1977; Mauck et al, 1978), although values for invertebrates tend to be one or more orders of magnitude lower. It should be noted that bioconcentration factors may be independent of the PCB concentration in the water (Defoe et al, 1978). In contrast to the bioconcentration factors demonstrated for PCBs, those for lindane reported in current literature, (see for example Thybaud and Caquet, 1991), for a range of aquatic organisms are much lower, reflecting the physicochemical properties of this compound. With the aquatic moss *Cinclidotus danubicus* bioaccumulation factors for hexachlorocyclohexane isomers ranged from 294 to 616 whilst for PCBs the value was found to be 4867 (Mouvet et al, 1985). Data on BCFs for invertebrates indicate that values are generally lower than for fish and are quite variable. Work by Saunders and Chandler (1972) found that bioconcentration factors of labelled Aroclor 1254 by aquatic invertebrates ranged from 2500 for *Pteronarcys dorsata* (stonefly) to 47 000 for the water flea *Daphnia magna* after an exposure period of four days. *Mytilus edulis* (common mussel) has been widely studied and bioconcentration values under laboratory conditions are available for a range of organic chemicals. The BCF factor for fenitrothion was 104, lindane was 154, while PCB and DDT were found to have BCFs of 15,650 and 23,650 respectively (Geyer et al, 1982).

Methodologies have been proposed for predicting the environmental fate of organic chemicals on the basis of certain physical / chemical parameters including partition coefficients (Neely, 1979; Maki, 1979). The solubility characteristics, bioconcentration factors and acute toxicities of Red List Organics are given in Table 3.2a.

Boryslawskyj et al (1987) demonstrated that in the freshwater bivalve
Sphaerium corneum (orb shell cockle) accumulation of contaminated particulate material is only a minor route of entry and contributes little to the overall body burden of the animal, with the primary route of dieldrin uptake being by direct partitioning of residues into lipoidal tissues. For an organism that accumulates the bulk of its residue burden by direct uptake from water, the proposal that the amount taken up is controlled by a factor closely related to the lipid/water partition coefficient for the residue seems to provide a semi-quantitative description of the net uptake process (Boyslawskyj et al 1987).

The lipophilic nature of organometallic compounds such as TBT is often invoked as an explanation for the high partition coefficients and hence bioavailability of such compounds to marine organisms. However, Langston et al (1987) concluded that lipid partitioning per se successively by higher members of the food-chain. Dichlorvos is one of the most volatile organophosphorus pesticides and is also rapidly degraded. Its rapid hydrolysis to form water soluble compounds means that it is not bioaccumulated or biomagnified.

2.7 Herbicides

Since the use of persistent organochlorine insecticides is now substantially restricted their occurrence in the environment is decreasing. Herbicides on the other hand are used in much larger quantities and so with their greater water solubility and thus potential to leach from sites of application they have been found to frequently exceed maximum acceptable concentrations in water.

Use of the triazine herbicides in substantial quantities both for agriculture and in non-agricultural uses (such as spraying railway embankments, roadside verges and in parks) has lead to their detection throughout the hydrological cycle, in rivers, lakes, drinking water supplies, groundwater and rainwater. Despite this, there seems to be little known of the mechanisms of bioaccumulation of herbicides, in contrast to the literature on accumulation of insecticide residues.

While the s-triazines are somewhat hydrophilic, with a water solubility in the mg/l range (Elleghausen et al, 1980) they must be considered as persistent compounds. Gunkel (1981) demonstrated that with atrazine, even under long term bioaccumulation conditions no degradation of accumulated residues occurred in fish. This confirmed investigations by Streit and Schwoerbel (1976) with *Ancylus fluviatilis* (river limpet) and by Böhm (1976) with algae. Mauck et al (1976) found that simazine residues in aquatic fauna can last in excess of a year, with bioaccumulation occurring in invertebrates.

Gunkel and Streit (1980) have demonstrated that the uptake of atrazine by the mollusc *Ancylus fluviatilis* (river limpet) and the white fish *Coregonus fera*, was rapid with water exchange as the main mechanism of atrazine accumulation and exchange. However, Macek et al, (1976) reported that atrazine does not accumulate in higher trophic levels and is relatively non toxic to invertebrates and fish.

Ambient concentrations as low as 20 μg atrazine/l have been associated with adverse effects on freshwater aquatic fauna, including benthic insects (Dewey 1986) and teleosts (Kettle et al 1987) although the effects were considered indirect. For example the richness of benthic insect species and total adult emergence declined significantly with atrazine addition, presumably by way of reduction in food supply of nonpredatory insects, and to some extent their macrophyte habitat. Bioaccumulation of atrazine from freshwater is limited and food chain biomagnification is negligible.

2.8 Organic Solvents

The published literature available on organic solvents is extremely limited and very little is known of the mechanisms of bioaccumulation. Oliver (1984) analysed the bioaccumulation of 15 different organochlorine compounds from contaminated sediments by oligochaete worms (*Limnodrilus hoffmeister* and *Tubifex tubifex*) and demonstrated low concentration factors (0.17) for compounds such as hexachlorobutadiene. This contrasts with a concentration factor of 2.6 for DDE. It is likely that the reason for the low concentration factors of organic solvents is due to their volatility which would also cause difficulties for laboratory studies on the bioaccumulation properties of these compounds.

2.9 Triorganotin Compounds

The use of biocides in antifouling coatings on boats and cage nets used in mariculture operations has resulted in tributyltin (TBT) contamination of coastal areas. The triorganotin compounds in the antifouling coatings are gradually released into the seawater where they act as toxicants towards marine fouling organisms. In particular, TBT contamination has been demonstrated in many harbours and marinas, where boats and yachts are concentrated (Bryan et al, 1986; Davies et al, 1987).

Organotins adsorb to particulate material and so the concentration in the water depends on the behaviour of the sediment. The levels of deposition, resuspension and mixing, for example, will have an influence on organotin concentrations (Ebdon et al, 1988). The sediments represent a significant source of contamination and for benthic organisms bioaccumulation of only a small proportion of this sediment burden may be significant.

TBT compounds are not as persistent as chlorinated hydrocarbons, and are not widely dispersed in the marine environment. There importance as an environmental issue was realised by the threat they posed to non-target organisms especially commercially important bivalves such as oysters. It appears that molluscs are particularly sensitive to TBT compounds (Langston et al, 1990).

As a consequence of this, paints containing TBT are now restricted to use on vessels exceeding 25m, under the Control of Pollution (Anti-fouling Paints) Regulations (1987).

In 1989, two years after the introduction of the ban, there were signs that sea water concentrations of organotins were declining (Langston et al, 1990). This decline will depend, however, on the extent to which TBT trapped in sediments is released and the rate at which it is degraded.

could not fully explain wide variations in concentration factors for benthic invertebrates. Species - specific characteristics including the ability to metabolise organotins, the mode of nutrition and contact with particulates may be more important in determining the bioaccumulation potential.

These relationships suggest that the overriding mechanism of uptake and retention is diffusion between the lipids of the organism and the surrounding aqueous environment.

Table 3.2a *Relationship of Solubility Characteristics of Red List Organics to Bioconcentration Factors and Acute Toxicities [Sources: Water Research Centre,1988; Biggar et al, 1966]*

Compound	Water Solubility	log K_{ow}	Maximum Reported BCF (in animals)	Acute Toxicity (96h LC_{50}) to Trout
DDT	3.1-3.4 µg/l @ 25°C	6.19	40000	1-126 µg/l
HCB	110 µg/l @ 24°C	6.18	3740	-
PCB	54 to 250 µg/l @ 25°C	6	47000	-
Trifluralin	24 mg/l	5.34	6000	10-40 µg/l
PCP	14 mg/l @ 20°C	5.01	475	-
Trichlorobenzene	4-49 mg/l (Depending on isomer)	4.1-4.3	5000	710 µg/l 24h
Triphenyltin Chloride	1-5.2 mg/l	4.1	-	-
Triphenyltin Acetate	9 mg/l @ 20°C	high	-	-
Triphenyltin Hydroxide	1.2 mg/l	3.93	-	0.015 mg/1 (Fry)
Lindane	17 mg/l @ 24°C	3.72	727	32 µg/l
Endosulfan	325 µg/l @ 22°C	3.60	2755	1.6 µg/l
Fenitrothion	14-30 mg/l @ 30°C	3.38	250	2 mg/l
Aldrin	105 µg/l @ 15°C	3.01	287500*	1.1 µg/l*
Azinphos-methyl	33 mg/l @ 20°C	2.99	-	4.3 µg/l
Malathion	145 mg/l @ 20°C	2.89	2.4	23 µg/l
Atrazine	33 mg/l	2.7	68	8.8 mg/l
Dieldrin	90 µg/l @ 15°C	2.6	287500*	1.1 µg/l
Simazine	5 mg/l	2.3	20	5 mg/l (24h)
Tributyltin Oxide	19.5 mg/l	2.29	-	15 µg/l
1,2-Dichlorethane	8.69 µg/l @ 20°C	1.76	21	225 mg/l
Dichlorvos	10 µg/l @ 20°C	1.4	-	170 µg/l (24h)
Endrin	130 µg/l @ 15°C	-	387500*	1.1 µg/l
HCBD	2 mg/l	-	17000	-

* Combined aldrin, dieldrin and endrin.

3.3 Importance of Chemical Speciation

Certain pollutants may exist in different species or forms in the marine environment and a particulate form of a toxicant will behave physically and biologically different from the dissolved form (Nelson and Donkin, 1985).

Cox (1971) found very little DDT in solution in seawater samples, with apparently 90% being bound to very small particles and unavailable to phytoplankton. Other organochlorines with low solubilities are likely to behave similarly. This has been confirmed for dieldrin (Dawson and Riley, 1977 and Rickard and Dulley, 1983) where elevated levels in a water body have been shown to be associated with high levels of suspended particulates.
Boryslawskyj et al (1987) showed that 35-45% of the total water burden of dieldrin may be bound to particulate matter in freshwaters.

The relatively simple use of the octanol-water partition coefficient to describe uptake can be modified by the presence of complex organic molecules, such as humics, in the water. Boehm and Quinn (1976) suggested that in the absence of Dissolved Organic Matter (DOM) organic contaminants are present in aggregations large enough to be removed from suspension by the filter feeding mechanisms of animals such as bivalves. Although physicochemical speciation can influence bioaccumulation, as can metabolism, solubility has such an overriding influence upon uptake and retention that simple models based on the octanol-water partition coefficient can still, in the opinion of Gosset et al (1983), be used. A simple one-component system like this has, however, its limitations and has sometimes failed to give useful predictions, for certain fish/pollutant combinations (Conner 1983).

3.4 Accumulation from Sediments

It has been well established that hydrophobic organic chemicals rapidly become associated with sediments and suspended particles (Bellar et al, 1980; Chiou et al, 1979, 1983; Karickhoff et al, 1979; Nau-Ritter et al, 1982; Zierath et al, 1980). Sediments contain significant levels of lipids making them sinks for hydrophobic compounds (Freed and Chiou, 1982). Bioaccumulation studies of sediment-sorbed organic chemicals have identified three probable pathways of chemical transfer.

- Interstitial water.
- Ingested particles (organic and inorganic).
- Direct contact with sediment particles.

The availability of sediment-sorbed trace organics is important since the sediment reservoir may constitute a primary source of contamination for benthic organisms (Knezovich et al, 1987). Fish and shellfish populations have been shown to accumulate chemicals from areas of known sediment contamination (for example Connor, 1984; Gosset et al, 1983). Degradation of pollutants is generally slower in sediment than in water overlying the sediment

(Rawn et al, 1982; Sharom and Solomon, 1981; Muir et al, 1985) and hence as the water column concentration of a contaminant decreases, its biological fate is increasingly likely to be related to the availability of the compound in the sediment. Courteny and Langston (1978) suggest that sediments may be the source of PCBs in the infauna only in less contaminated areas.

The bioavailability of sediment-sorbed chemicals may be influenced by a number of factors. One of the most important is sediment type with many researchers reporting an inverse relationship between chemical availability and sediment organic carbon content (for example Adams et al, 1983; Larsson 1984; Muir et al, 1985; Oliver, 1984). Duinker et al, (1983) claimed that the content of organochlorines in sediments appeared to be determined primarily by the grain size distribution with a significant positive correlation established between the fraction of particles <50 μm and levels of total PCB, DDD and DDE. With fine grained sediments there is an increased surface area available for adsorption and a reduced volume of interstitial water. No significant correlations were found with the less hydrophobic HCB, alpha and gamma HCH and dieldrin.

Interstitial water plays an important role in the bioavailability of chemicals. It is formed due to the entrainment of water during the sedimentation process and is essentially isolated from the water column. The desorption of sediment-sorbed organic chemicals is mediated by interstitial water. As infaunal organisms are in direct contact with interstitial water, the concentrations of desorbed chemicals contained in this compartment will directly affect bioavailability (Knezovich et al, 1987). A number of studies have found that bioconcentration is primarily due to partition between the organism and interstitial water. Markwell et al, (1989), have produced an equation allowing interstitial water concentration to be calculated from sediment concentration, hence enabling a bioconcentration factor to be determined.

$$\log K_B = 1.1 \log K_{ow} - 1.0$$

where

K_B = bioconcentration factor
K_{ow} = octanol-water partition coefficient.

The bioaccumulation of lipophilic compounds by oligochaetes cannot be satisfactorily interpreted on the basis of concentration of compounds in the sediments (Connell, 1988). If, however, the corresponding interstitial water concentrations are used, then a set of significant relationships can be obtained. These studies do, therefore, suggest that bioconcentration from interstitial water is likely to occur in oligochaetes.

Chemical transfer of sediment-sorbed organic pollutants can occur by the ingestion of both organic and inorganic particles. The importance attached to this pathway is varied and while some studies have concluded that ingestion is an important route of entry (Fowler, 1978; Langston, 1978; Gosset et al, 1983), some have suggested that it may contribute to the total body burden (Muir et al, 1985; Elder et al, 1979), while still others have dismissed it claiming that the main uptake occurs via other routes (Larsson, 1984; Knezovich et al, 1987; Connell, 1988). It has been suggested that these discrepancies may be due in part to differences in the interpretation of results as well as differences in methodology.

There is only limited information available about the third possible pathway of chemical transfer, by direct contact with sediment particles, as it has not been extensively studied. However, direct absorption to the body wall or exoskeleton has been demonstrated with crustaceous chitin and chitosan (Davar and Wightman, 1981; Kemp and Wightman, 1981) and absorption through the annelid cuticle, has also been observed (Lord et al, 1980).

Once contaminants have become associated with sediments, they may become available to food webs by a number of pathways. These principally are resuspension, diffusion from the sediment into the water column, ingestion, absorption or adsorption. Fowler et al (1978) suggested that approximately 99% of total uptake of PCBs by polychaetes would be derived from sediments. Since benthic organisms are generally able to accumulate chemicals from particulate and interstitial components of sediments as well as the water column they may be at a greater risk than pelagic organisms (Swartz and Lee, 1980). Shaw and Connell (1982) conclude that for some estuarine benthic organisms, sediment concentrations were the major influence on body concentrations of PCBs and a direct relationship between body concentration and the log of sediment concentrations was established.

3.5 Predator Prey Relationships

When aquatic organisms are exposed to organic chemical contaminants, uptake may occur by a variety of routes, some of which may be difficult to distinguish. Uptake from water, appears to be of overriding importance, while the significance of food varies according to the literature selected. Cosset et al (1983) claim that the main route of contaminant uptake is via food supply while Ellgehausen et al (1980) take the opposite view with food being far less important than uptake from water.

It has been reported that while data may not support the idea that food is the main source of contamination, it does need some consideration since fish preying on detritus feeders do have higher levels of pollutants (Bjerk and Brevik, 1980).

Some studies have shown that the main route of uptake of organic contaminants in marine organisms appears to be via their food (Young et al, 1980; Schafer et al, 1982) and as such uptake may depend largely upon the availability of contaminants to the base of the foodweb. Absorption from the gastrointestinal tract is probably independent of the octanol : water partition coefficient of contaminants since organisms are designed to absorb both lipophilic and hydrophillic substances. Food may play a more important role in the amounts of organic chemicals accumulated at higher trophic levels. This may be due to differences that the predator and prey may show in diet, size, lipid content and age, and should be considered in predator-prey relationships. In some cases organisms lower down the food chain may act as a means of maintaining contaminant levels in the food chain through remobilising trace organic residues present in sediments (Elder et al, 1979).

Trophic level as a factor affecting indicator organism reliability and the concept of food as a source of contamination are discussed further in Section 5.10.

4 BIOACCUMULATION COMPARED WITH OTHER APPROACHES

4.1 Analysis of Water

4.1.1 Advantages

- Measurements of Red List trace organic concentrations in water relate directly to current water quality standards.

- If waters are relatively clean and free from interfering substances, then with current instrumentation analysis is relatively easy and sensitivity can be good.

4.1.2 Disadvantages

The determination of Red List trace organics in water samples can pose a number of problems:

Freshwaters

- Certain compounds at sub-$\mu g l^{-1}$ concentrations may be difficult to detect in waters carrying interfering substances. In such areas analysis with confidence is likely to necessitate expensive instrumentation such as gas chromatography linked to mass spectrometry.

- In rivers intermittent industrial discharges can result in "pulses" of a contaminant being released such that at a given time of sampling the concentration in the water can be much higher a few kilometres downstream from the point of discharge than immediately below it. This is a major drawback for the precise location of the sources of pollutants.

- If water sampling does not take place at the time of passage of an intermittent discharge or a "pulse" of more diffuse pollution passing downstream, it is quite likely that detection of the pollutant will be missed altogether.

Estuarine Waters

- The analysis is complex, laborious and can involve the use of expensive equipment for collecting samples (for example multiple samples, time integrated etc).

- The inherent variability at sites due to factors such as the amount of freshwater run-off, currents, tides, seasonal effects, sampling depth, climatic influences, intermittency in effluent discharge, all necessitate at least multiple sampling.

- Difficulty in separating meaningful fractions in which organochlorine compounds may be associated eg. dissolved, particulate (organic, inorganic) etc.

4.2 Analysis of Sediments

4.2.1 Advantages

- In some localities sample collection can be relatively easy (small shallow streams and rivers).

4.2.2 Disadvantages

The limitations in the use of sediment analysis for detecting and monitoring Red List trace organics in rivers and estuaries include the following:

- Errors can be introduced because of local variation in sedimentation rates of particulates and because of variable amounts of organic matter present.

- Sediments can be transported long distances in rivers making the detection of a polluting discharge or the localization of the main zone of impact almost impossible.

- Sediments can also be moved around within estuaries as a result of the dynamics of tides, currents and wave action.

- Problems and complexities when dealing with different particle size fractions.

- Difficulties in interpreting results in terms of potential biological effects or bioavailability.

4.3 Bioaccumulation

4.3.1 Advantages

Use of biological material offers several advantages over other approaches for the assessment and monitoring of trace organic contamination and these include the following:

- Contamination levels are integrated over an extended period of time and are therefore more representative of environmental quality than are instantaneous samples of water and sediment. Sampling frequency can therefore be reduced without sacrificing accuracy resulting in considerable savings in terms of time, effort and costs.

- Results generally reflect the biologically available concentrations of the organic pollutant rather than merely its presence at a particular concentration in the environment.

- Bioaccumulation usually provides tissue concentrations of trace organics which can be detected with relative ease using readily available techniques / procedures.

If the attributes necessary for selection as a biological monitor are adhered to (see Section 4.3) then bioaccumulation as an approach can be both very useful and effective.

4.3.2 Disadvantages

Use of higher organisms within the food chain can bring some potential drawbacks to the approach such as:

- difficulty in relating results to actual water concentrations;

- potential problems in interpreting the data quantitatively due to biological variability (see Section 5.4, 5.5 and 5.6);

- the possible interference of extraneous parameters with the uptake of pollutants by biota (see Section 5.7 and 5.8);

- difficulty in sampling, as there may be the need for sophisticated trapping equipment which can be time consuming and expensive;

- some organisms are not necessarily tolerant of high levels of toxicant (see Section 5.9).

4.3.3 General comments

There is an attraction and some clear advantages in using simpler organisms, in particular attached plants (algae, mosses) because the complexity in many of the potential disadvantages listed above are substantially reduced. For example:

- Due to the high levels of accumulation found in simple plants, for example, concentration factors of 3000 for DDT in *Cladophora* (Meeks, 1968), 10000 for PCBs and 300 for gamma-HCH in mosses (Frisque et al 1983), measurement of the levels in such organisms certainly increases the sensitivity of detection.

- The collection of suitable plant materials provides an easy method for people without access to sophisticated water sampling or higher organism sampling facilities. Being sedentary and easily recognizable plants can be easily collected. Samples of moss can, for example, be collected by anyone who suspects that contamination may occur at a site.

- Plant "monitors" give an integrated record of pollution within a particular system. This may be especially important where pollution is intermittent.

- Plants can be harvested after a pulse of contaminated water has passed downstream so it should be possible to pinpoint pollution, hours, or perhaps even days after it has occurred.

- It is reasonable to assume that organochlorine accumulation by a plant gives a better indication of the fraction of the residue in the environment which is likely to affect the biota (ie. directly available from the water) than most types of direct chemical analysis.

- The use of algae and mosses, in particular reflects the ambient concentrations of a pollutant in the water and is not complicated by additional sources, either directly from sediments, or through predator / prey relationships.

- Plants are relatively simple organisms and there are no complications associated with movement, feeding habits, life cycles (spawning / reproduction) and migrations.

- Plants such as aquatic mosses can be easily transplanted to a contaminated site (for example attached to boulders) and used for monitoring purposes (Mouvet et al 1985).

As with all approaches there are inevitably a number of potential complications in using attached algae or mosses. In particular, the seasonal abundance of certain algal species may complicate matters and for all attached plants the potential effects of flooding (scouring and removal of material) may also impact on their effective use in certain situations.

4.4 Considerations in using Bioaccumulation

The most important considerations in a bioaccumulation monitoring programme are:

- the standardisation of techniques throughout the area of programme implementation;

- the appropriate choice of indicator species.

The use of bioaccumulation techniques in a monitoring programme for trace organic compounds requires careful planning and execution in the selection of indicator species, sampling procedures and the methods of analysis. As noted by Phillips (1978), the relative abundance of pollutants at different study locations may be accurately predicted by the use of biological indicators only if such precautions are taken.

Many species are inappropriate as indicators because they are too scarce, too difficult to sample or because they regulate the body-burden in some way. Consequently, it is generally recognised that no one species is capable of acting as a universal indicator.

4.5 Attributes of Indicator Organisms

The required attributes for an organism to act as a biological monitor of aquatic pollutants were developed primarily in studies of marine pollution by trace metals, but may equally be applied to studies of organic contamination in the aquatic environment. The attributes were first outlined by Butler et al (1971) and were subsequently added to by Haug et al (1974) and Phillips (1976, 1978). They include:

(a) The organism should accumulate the pollutant without being killed by the levels encountered in the environment.

(b) The organism should be sedentary in order to be representative of the study area.

(c) The organism should be abundant throughout the study area and preferably have a widespread distribution to facilitate comparison between areas.

(d) The organism should be sufficiently long-lived to allow the sampling of more than one year class, if desired.

(e) The organism should be of reasonable size, giving adequate tissue for analysis.

(f) The organism should be easy to sample and hardy enough to survive in the laboratory, allowing defecation before analysis (if desired) and laboratory studies of pollutant uptake and loss.

(g) The organism should tolerate brackish water (for marine or estuarine system studies).

(h) All organisms of a given species used in a survey should exhibit the same correlation between their pollutant content and the average pollutant concentration in the surrounding water, at all locations studied, under all conditions.

Table 4.5a Characteristics of Indicator Organisms Related to Criteria of Phillips (1978) Criteria of Bioindicator

Organism	a	b	c	d	e	f	g	h	Reference
Algae									Vance and Drummond, 1969
Aquatic bryophytes									Mouvet *et al* 1985
Macrophytes									Gobas *et al*, 1991
Fontinalis antepyretica									Morrison and Wells 1981
Leeches									Metcalfe *et al* 1984
Hydrobia jenkinsi									Brown 1980
Nereis virens									Goerke and Webster, 1990
Nereis diversicolor									Goerke and Webster al 1990
Sphaerium corneum									Boryslawskyj *et al*, 1987
Freshwater mussel (Unionidae)									Bedford *et al* 1973
Elliptio complanta									Metcalfe and Hayton, 1989
Anodonta piscinalis									Herve *et al*, 1988
Bivalve molluscs									Bayne, 1989
Bivalve molluscs									Langston, 1978
Mussels									Geyer *et al*, 1982
Mytilus edulis									Tanabe *et al*, 1987
Mytilus edulis									Goldberg, 1984
Mytilus edulis									Phillips, 1978
Buccinum undatum									Knickmeyer and Steinhart, 1989
Nucella lapillus									Davies *et al*, 1987
Eel									Rickard and Dulley, 1983
Plactichthys flesus									Rickard and Dulley, 1983
Limanda limanda									Büther 1990

The criteria set out above have been followed to varying extents in the design and implementation of monitoring programmes. Table 4.3a illustrates the characteristics and reasons stated by the authors of the programmes/research that influenced them in their choice of bioindicator organism. It only includes those references where mention of the characteristics were given.

From Table 4.3a it can be seen that the extent to which most authors follow Phillips' (1978) criteria is inconsistent. Whilst many seem to consider the fact that the organism should accumulate environmental levels of pollutant without being killed as being important, it does not appear that any consideration is given to the final criterion, that the organism of a given species should exhibit some correlation between their pollutant content and the average pollutant concentration in the surrounding water at all locations studied, under all conditions. However, no other reference to this was found throughout the current literature review. The other main considerations appear to be that the organism should be abundant, and sedentary.

It should be noted that in Table 4.5a reasons given by the authors for their choice of organism are cited. The bioindicators may possess other characteristics that would recommend their use but these, however, were not mentioned in the literature.

4.6 Species Selection

Different types of organism will reflect levels of organic contaminants in different ways according to the pathways by which accumulation takes place. This is particularly important in the case of the organochlorine compounds since these exhibit a high affinity for both inorganic and organic particulate material and a low solubility in water (Phillips, 1978).

Macroalgae have been more frequently used for trace metal pollution than for organic contaminants. The reason appears to be that organochlorines in particular exhibit higher association with particulates than do most trace metals and these organisms respond mainly to pollutants in solution. Phillips (1980) has suggested that for pollutants occurring mainly in solution it may be preferable to use a primary producer, thus avoiding food chain effects. However, it has been suggested that in polluted situations, the solubility maxima for some compounds may be exceeded, producing a 'saturation effect' above which the levels of contaminants in macroalgae will not respond.

There are difficulties in comparing species that accumulate pollutants in different ways. Thus macroalgae, which respond only to soluble constituents may be expected to produce different profiles of contamination from those produced by a filter-feeding mollusc which is responsive to organic contaminant loads in solution, in water, as well as contaminants in association with inorganic particulate material and in food (Phillips, 1978).

The profiles in the study of organochlorines may also vary since the ratio of soluble to particulate associated organochlorine may differ from location to location. This ratio may vary in accordance with local conditions, for example, the type of organic material present in the water column. Since benthic organisms are generally able to accumulate chemicals from the particulate and interstitial components of sediments as well as from the water column (Swartz and Lee, 1980), they may be able to bioaccumulate a greater body burden. The extent to which the pathways contribute to the total accumulated body burden of an organism varies according to the individual species involved and also with the ambient environmental conditions.

Teleosts respond to both dissolved pollutants and to those originating from contaminated food. There are few species, however, that act as accurate bio-indicators since only a small number are sedentary and they are relatively difficult to sample. Their widespread occurrence, relevance to public health issues, relatively high lipid content and coincidence with NSTF monitoring work means, however, that they are a valuable part of a monitoring scheme.

The use of multiple indicators in any one study should lead to a more complete understanding of the pollution of the area, especially if the species used respond to different portions of the total pollutant load in an ecosystem (Phillips, 1977a).

5 FACTORS AFFECTING INDICATOR ORGANISM RELIABILITY

5.1 Introduction

In this section the various factors that affect the observed concentrations of organic contaminants in biota are reviewed. Recognition of these factors, and means of reducing their influence, is of central importance to any monitoring programme.

The bulk of the available literature and the two major reviews on the use of biological indicators for organic contaminants (Phillips 1978; 1980) have only been concerned with organochlorine compounds and thus much of the following section relates to this group of compounds. Observations made with regard to these compounds, however, could be expected to apply to other hydrophobic organic contaminants.

Many of the indicator surveys performed to date have not considered external variables in enough detail to allow any accurate estimation of pollution conditions in any study location.

5.2 Accumulation, Degradation and Elimination

In addition to the attributes of the organism itself, the time integration of different pollutants should be considered in both the selection of indicator species and also the intervals between successive samples in continuous monitoring surveys (Phillips 1978). The rates of uptake and excretion of a pollutant by an organism determine to a great extent, the degree of time-integration of ambient pollutant levels exhibited by that organism.

Different species exhibit widely divergent rates of uptake and excretion for any one contaminant, and the variation depends little on taxonomy; thus closely related species may not necessarily exhibit similar pollutant kinetics. Butler (1971) has reported considerable differences between the uptake and flushing rates for seven pesticides in *Mya arenaria* (soft clam) and in *Mercenaria mercenaria* (hard clam). In freshwater salmonids, the excretion rates of DDT in *Salmo salar* (Atlantic salmon) and *Salmo gairdneri* (rainbow trout) appear quite similar (Reed 1966; Sprague et al 1971) but (*Salvelinus fontinalis* (American brooktrout) excretes DDT much faster (Cole et al 1967).

Any one species will exhibit unique kinetics for each pollutant. These differences are often noted for pollutants of very similar chemical structure such as DDT or DDE, or different isomers of PCBs. Thus, each species-pollutant pairing is unique in terms of pollutant kinetics. The frequency of sampling biota in monitoring surveys should be defined by the kinetics of contaminants in the chosen organism(s). This parameter may be of over-riding importance for organochlorine monitoring since these compounds exhibit very short half-lives in some species.

Present knowledge of PCB kinetics in bivalves suggests that 'time-bulking' of samples (combining multiple samples of a bioindicator species taken at frequent intervals from a single location) should be considered for future monitoring studies of PCBs using bivalves, since a more accurate picture of average contamination conditions would be provided (Tanabe et al, 1987).

The higher flux of organochlorines through biota when compared with trace metals as for example in *Crassostrea virginica* (eastern oyster) (Brodtmann 1970) necessitates more frequent sampling of indicator organisms for monitoring studies on organochlorines (Butler 1966, 1969a). Rapid elimination of DDT from *Crassostrea virginica* (eastern oyster) required regular sampling (at 30-day intervals) of this species as part of the National Monitoring Program for organochlorines in the USA (Butler 1973). The rates of uptake and elimination of pesticides by teleosts are generally slower than those of bivalve molluscs (Butler 1971). Kinetics and half lives are highly relevant to the design of monitoring surveys which employ bioindicators to reveal geographical or temporal variations in contaminant abundance. Fluctuations in environmental concentrations of pollutants and differences between the rates of uptake and loss of contaminants will affect the net residue concentration at any given time. In addition rates of accumulation and more particularly, equilibrium concentrations of some substances depend much on the condition of the accumulator.

If a continuous record of pollutant availability at any study site is required, the frequency of sampling must be adjusted to account for the time-integration capacity of the species used with respect to the particular pollutant measured. Phillips (1980) stated that the great divergence in physicochemical properties of individual PCB components warranted the treatment of each component as a pollutant in its own right, and several authors (for example Neely et al, 1974; Ernst, 1980; Mackay, 1982) have reported correlations between PCB concentrations and physicochemical properties of individual PCB components.

5.3 Effect of Body Lipid

As has been mentioned before trace organic pollutants (in particular the organochlorine compounds), although only slightly soluble in water, are extremely soluble in lipid or fatty tissues of biota. This property accounts for the high biological affinity of organochlorines as well as their stability and persistence in aquatic biota (Risebrough et al, 1967). The amount of lipid present in an organism is undoubtedly the most important extraneous parameter determining the uptake and retention of organochlorines. The only factor of greater importance is the actual availability of these compounds in the ambient environment. The lipid levels are responsible for many of the reported differences in organochlorine concentrations between species or between individuals of any one species, and they also contribute (at least in same cases) to the variations in pollutant levels in biota with age, season and possibly other parameters.

Several parameters are known to affect the amount of total lipids in aquatic biota. Of these, the most important is season and the associated variation in organism lipid content related to the sexual cycle and to the water temperature (Phillips 1980). In general, the lipid content of aquatic biota increases prior to and during gonad development, with decreases being evident during and after spawning.

Individual animals of any one species at any one location may vary greatly in the percentage lipid present in the body. This may be due in part to factors such as age, migration and sex. Consequently this is an important potentially disruptive factor in an indicator survey for organic contaminants. Furthermore, there is often an increase in body lipids after exposure of teleosts to sublethal levels of DDT, dieldrin or endrin (Macek et al, 1970; Grant 1976) which may further complicate interpretation. In addition, sublethal growth effects due to organochlorines may be seen in both teleosts and bivalve molluscs. Data concerning the effects of lipid on species or tissue differences in organochlorine concentrations amongst organisms other than teleosts are sparse. However, correlations have been found between tissue distributions of pollutants and the lipid content of tissues (Ernst et al, 1976; Geyer et al, 1982; Holden 1962; Duursma et al, 1986).

Several authors have reported that the variation in the concentration of organochlorines in individuals of one species at any one location is less on a lipid weight basis than on a wet weight basis. Duinker et al (1983), found that concentrations of PCBs on a lipid basis did not increase with size or age in samples of the tellin, *Macoma* and the shrimp, *Crangon* originating from the same sampling area. Likewise Geyer et al (1982) found that the standard deviation among individuals was greatly reduced by using a lipid weight basis for calculating bioaccumulation factors for mussels. Thus the use of a lipid weight basis for organic contaminant concentrations in biota eliminates the differential effects of lipid, although certain assumptions are inherent in this method (Mackay, 1982), as detailed below.

(i) Provided degradation and other processes act in an equivalent manner, lipophilic compounds will be distributed within an organism in proportion to the lipid content of the tissues involved.

(ii) Provided exposure conditions are equal, the total concentration of a lipophilic contaminant in a population of individuals of equal physiological and other characteristics, will be proportional to the total lipid concentrations in the individual.

The available data are usually in accord with these generalisations, both in the field and in laboratory studies (Connell, 1988). The method does, however, presuppose that the uptake from water is far more important than that from food in the overall accumulation of organochlorines by biota. If this model is valid, bioaccumulation factors determined for one indicator organism may be used to calculate the bioaccumulation factor in other organisms, if the lipid content of the species involved is known (van der Oost, 1988).

There are authors, however, who state that these relationships are not necessarily valid and that the pollutant levels of the total animal are not determined by fat content but mainly by other factors such as reproduction and equilibrium partitioning in the water column (Hummel et al, 1990; Ernst and Goerke, 1974; Gunkel, 1979a; Hesig-Gunkel and Gunkel, 1981).

It seems that while lipid content is not ideal for estimating organochlorine concentrations since it does not take into account variations in lipid composition (and hence the precise affinity of contaminants for the lipid pool), it is generally an acceptable approximation (Phillips and Segar, 1986).

5.4 Species and Tissue Differences in Organic Contaminant Concentration

5.4.1 Introduction

Important differences have been reported in levels of organic contaminants between different species and also between different tissues in any one species. These variations should be considered in the design and implementation of a monitoring programme employing bio-indicator organisms.

5.4.2 Variation Between Species

Differences in the levels of organochlorine concentrations between different teleost species have been ascribed by many authors to differences in the lipid contents of these species (see for example Duffey and O'Connell, 1968; Jensen et al 1969, 1972; Reinert 1970). Some authors have been, however, unable to show any correlations between the lipid content of the fish species and their concentration of organochlorines (Hattula et al 1978a; Henderson et al 1969, 1971). These results may in part be due to the combined and interactive effects of other factors, such as season and fish size. An important factor that may cause variations can be found in the ability of an organism to metabolise the pollutants, and the presence of the enzymes to carry out this function. Fish in general have the best potential for metabolism among aquatic animals (Schüürmann and Klein, 1988) and are able to metabolise compounds to different extents, by a range of transformation reactions (Lech and Bend, 1980). The activities of the enzymes involved vary widely within, and between species, with season and with temperature (Pyysalo et al, 1981; Neff, 1979; James et al, 1977). The levels of these enzymes can reach high levels, especially when in an induced state (Stegeman et al, 1981; Connor, 1983). Bivalve molluscs do not appear to be able to metabolise organic contaminants with the same facility as some crustaceans and fish (Neff et al, 1976; Goldberg, 1986) and so the levels which are accumulated depend upon factors such as the lipid-water partition coefficient and the amount of lipid in the organism (Bryan, 1979). Broadly, the activities of the microsomal mono-oxygenases present follow the order molluscs, <polychaetes/crustaceans, <fish (Connor, 1983) (see also Section 5.2).

Differences may be seen in the concentrations of organic contaminants in species within the same group, for example, *Mercenaria mercenaria* (hard clam) exhibits different bioconcentration factors when compared with *Mya arenaria* (soft clam) (Hawker and Connell, 1986). There may be differences in the oxygen uptake mechanism involving different water ventilation processes that would lead to different uptake rate constants for different aquatic organisms (Connell, 1988). *Crassostrea virginica* (eastern oyster) accumulates twice as much DDT (Butler, 1973) compared with *Mercenaria mercenaria* (hard clams), but the oysters feed at a faster rate (Tenore et al 1973). These variations have been observed in other groups. Elimination kinetics of single PCB components showed that *Nereis diversicolor* (common ragworm) possesses mechanisms of biotransformation which differ from those of *Nereis virens* (king ragworm) and this may appear unusual for two such closely related species. Neuhoff (1979) demonstrated, however, that they have very different growth efficiencies which would indicate differences in metabolism.

5.4.3 Variation Between Tissues

Variation in the concentration of organochlorines within different tissues of a single fish species has been correlated with the lipid content of the tissue. For example, Holden (1962) reported that the distribution of ^{14}C-DDT among the tissues of *Salmo trutta* (brown trout) exposed to this compound experimentally was directly related to the lipid content of the tissues. Similar conclusions concerning the relationship between tissue lipid content and the ability to sequester organochlorines were reported for lindane (gamma-BHC) in *Salmo gairdneri* (rainbow trout) and *Rutilus rutilus* (roach) (Tooby and Durbin, 1975). Some information concerning the effect of lipid on organochlorine concentrations found in different tissues of finfish is also available from studies of fish taken directly from the environment. Linko et al (1974) reported similar results for *Esox lucius* (pike) from the Baltic Sea when compared with those for *Trigla lucerna* (gurnard) reported by Ernst et al (1976). The tissue distribution of DDT, its metabolites and PCBs in three species of finfish and *Chlamys opercularis*, (scallop) taken from the English Channel, was found to be closely correlated with the lipid content of the tissue (Ernst et al, 1976).

The tissue dependence of organochlorine concentration and its correlation with lipid contents of the tissues reaches its extreme in the axial muscle of some finfish. The correlation of muscle lipid profiles in the axial muscle with variations in organochlorine concentration in finfish was first reported by Reinert (1969) for *Oncorhynchus kisutch* (coho salmon). Rickard and Dulley (1983) found that in *Platichthys flesus* (flounder), the concentration of lindane was greatest in the liver by an order of magnitude. The levels in liver of *Osmerus eperlanus* (smelt) were ten times those found in the flesh and this was seen in most fish samples for different chemicals. The total residue is generally determined however by the residue in skeletal muscle since these parts represent a large portion (in most species, 90%) of the total weight of the fish (Gunkel, 1981; Wharfe and van den Broek, 1978). Such studies have not been restricted to finfish and similar results have been found with mussels (Bedford et al, 1973; Ernst, 1977) since the pollutant concentration was distributed according to lipid content, and was much higher in the viscera than in muscle. This pattern was also found in *Mercenaria mercenaria* (hard clam) (Courtney and Denton, 1976), *Lymnaea palustris*, (marsh snail) (Thybaud and Caquet, 1991) and *Ancylus fluviatilis* (river limpet) (Streit, 1979).

5.4.4 Conclusions

Differences in the concentrations of trace organics, in particular organochlorines between different species demonstrate the importance of metabolic enzymes, variations in uptake rates and lipid levels. Relationships should only be applied with caution since even closely related species have been shown to differ markedly in their bioconcentration behaviour.

Since organic contaminants in different tissues have generally been correlated with the lipid content of the tissue, this has important repercussions for tissue sampling practices in monitoring surveys. Subsampling procedures for a particular tissue should be considered very carefully and standardised in such programmes (Phillips, 1980).

Most authors report some residual differences in the concentrations of organochlorines found in different tissues of a single species or in different species, even on a lipid weight basis (Phillips, 1978). This indicates that lipid levels may not be the only determining factor for the observed differences but there is the possibility of different lipid types with different efficiencies of pollutant sequestration, in addition to possible variations in uptake and excretion rates.

5.5 Effects of Organism Age, Size (or Weight) and Length

5.5.1 Introduction

Concentrations of organic contaminants in bioindicators vary with age or size (Boyden, 1977; Phillips, 1980) and these variations may play an important role in the total levels of pollutant accumulated.

Data concerning the relationship between these factors and the concentrations of organochlorines present in biota are far more abundant for teleosts than for any other indicator type (Phillips, 1978).

5.5.2 Variation with Age

The mechanism producing age dependence of trace organics especially the organochlorines is commonly found to be the change in lipid content which accompanies ageing in some species (Phillips, 1980). It is also possible that with age there could be changes in the relative amounts of different lipid types that could lead to altered affinities of the total lipid pool for organochlorines.

Changes in the total lipid content of biota with age are best documented for finfish, particularly salmonids, although they have been observed in others such as *Anguilla anguilla* (European eel) (Lovern, 1938) and *Gadus morhua* (cod) (Jangaard et al 1967a,b). It thus might be expected for increases in body lipids to occur synchronous to the increased body lipids at any given exposure level. This will be reflected in an age-dependent increase in the concentration of organochlorines based on wet weights, whilst concentrations based on lipid weights would not be expected to change (Phillips, 1980). The general increase of lipids as a percentage of total body weight in older organisms is not the only factor contributing to increases in concentrations of organochlorines seen in some species with increased age or size. There is also the possibility of uptake rates varying in teleosts of different ages or sizes but of the same species.

Very little information exists concerning the variation in concentrations of organochlorines with age (or age-dependent parameters) in organisms other than finfish. Bivalve molluscs, which have been quite extensively studied as indicators of organochlorines, do not appear to exhibit age-dependent changes in lipid contents (Galtsoff, 1964; Giese, 1969; Bayne, 1976) and the same is true for gastropod and cephalopod molluscs (Giese, 1969). Seasonal variations in these organisms far outweigh any effects of age. No study to date is known in which convincing data have shown age dependence of organochlorines in molluscs (Phillips 1980). There is evidence, however, that age can play a role in short term exposure to TBT. Gibbs et al (1987) found that under these conditions, the degree of imposex in immature *Nucella lapillus*, (common dogwhelk) females that have yet to breed (individuals 12 - 18 months old) gave the greatest potential for monitoring purposes. Although some authors maintain that age based differences of organochlorine concentrations in teleosts are based on lipid changes with age, other workers have shown significant age-concentration variation even on a lipid weight basis. Interference by other factors such as location, season or migration may confuse the situation still further (see Section 5.5).

Rickard and Dulley (1983) found that for alpha-HCH and lindane levels in fish sampled from the tidal Thames, the concentrations in *Platichthys flesus* (flounder) were low in young age groups, while only 27% of fish over 2 years contained HCH below the detection limit used by MAFF in their Aquatic Environment Monitoring Reports. This is consistent with the fact that these compounds are less lipophilic than most other organochlorine compounds and hence their tissue levels show more age dependence than lipid content dependence. The concentrations of HCH in *Solea solea* (sole) showed high levels in the period of greatest growth rates, ie 1-2 years with levels dropping in the 2-3 year range. Endrin levels in *Solea solea* (sole) are at a maximum in the 1 + year group and were found to coincide with a high lipid content and then reduce with age. This is partly due to the fishes' ability to mobilise and store lipid-soluble products in their liver, and also because older fish are more likely to have spent more time in the cleaner waters of the southern North Sea. They concluded that the distribution of persistent organochlorine chemicals in fish tissues is more dependant upon fat levels of the organism than its age.

Phillips (1978) noted that there was an age-related variation in organochlorine residues which could not always be accounted for by changes in lipid content. He concluded that it must be age or size dependent and that no generalisations about size as a cause of indicator variability could be made. He stated further that the age of organisms sampled should be considered as a 'potential interfering factor' in indicator surveys especially when using fish.

5.5.3 Variation with Size or Weight

Animal size is an important parameter in uptake and several authors have found that the toxicity of pesticides is size dependent in fish (Pickering et al 1962; Mount, 1962 and Tooby and Durbin 1975). These and similar studies indicate that pesticides in solution are more toxic to small rather than large individuals of a given species. This may be related to reports describing greater uptake rates for smaller individuals (Murphy, 1971; Murphy and Murphy, 1971; McLeese et al, 1980; Connell, 1988).

Although small fish may accumulate DDT more rapidly (per unit weight) than larger individuals, at equilibrium the latter may exhibit a higher concentration of DDT (based on wet weights) because of the greater lipid pools (Phillips, 1980). The observed phenomenon could be the result of greater relative surface area or metabolism among smaller animals. A large surface to volume ratio increases the importance of uptake by dermal absorption (Bjerk and Brevik 1980). The gill area: body weight ratio changes with fish size (Connell 1988) and this suggests that small fish may accumulate lipophilic compounds at a more rapid rate. There have been mechanisms suggested where the surface area: body weight ratio of organisms may be an important determinant of uptake characteristics (Kenaga 1972; Harding 1977). This characteristic was found not to be significant for different sizes of zooplankton if the concentration was expressed relative to the dry weight of the organism (Harding and Vass, 1978).

Although data on invertebrates are sparse, the faster uptake of organochlorines in smaller individuals compared with larger animals has been observed for the uptake of methoxychlor by *Cancer magister* (marine crab). Wildish and Zitko (1971) suggest that PCB uptake from seawater by *Gammarus oceanicus* (marine shrimp) is dependent on the total body surface area of the organism. In contrast studies of *Chlamys opercularis* (scallop) revealed no weight dependence of organochlorine concentrations (Ernst et al, 1976). Size-dependence of contaminant concentrations, however, is rarely observed and should not be considered universal (Phillips, 1978).

In Swedish waters, the ratio of DDT to DDE approximates 1:1 in axial muscle tissues of fish such as *Esox lucius* (pike), or *Gadus morhua* (cod), both of which contain about 1% lipid in axial muscle tissues (Jensen et al, 1972, 1975). In contrast, *Anguilla anguilla* (eel), which contains 10-20% lipids in muscle, exhibits a much higher ratio of DDT to DDE, approximately 2:1 (Jensen et al, 1975). The authors suggested that fish with relatively low lipid contents turn over the lipid pool much faster than those with high amounts of lipid, thus freeing sequestered DDT for possible dehydrochlorination.

The effect that lipid content has on the rate of DDT dehydrochlorination to more polar metabolites was found in *Salmo salar* (Atlantic salmon) (Addison and Zinck 1977); older fish which generally contain greater amounts of lipid, should metabolise DDT at slower rates. This may have a bearing on the retention of DDT or its metabolites.

Reports which include information on the lipid content of freshwater fish, as well as their organochlorine concentrations, permit some estimate of the contribution of lipid changes to age dependence of organochlorines (Anderson and Everhart 1966; 1970). Whilst some of these data show that concentrations of sigma DDT and / or its metabolites, calculated on a wet weight basis, generally increase with age of *Salmo salar* (Atlantic salmon), age was certainly not the only parameter involved in the determination of DDT levels in these fish. Both the condition of the fish (i.e. the amount of body fat present) and the sex of the fish thus appeared to alter sigma DDT accumulation and both gave rise to a large amount in variation of any given year class. These authors concluded that such individual variability made it almost impossible to elucidate the actual sigma DDT levels present in the population using random sampling methods. They therefore suggest that selective sampling according to fish age, fat content and sex was far more likely to provide an accurate estimate of the sigma DDT levels present in this species. Whilst *Salmo salar* (Atlantic salmon) may be more variable than most other species in terms of individual lipid contents, this suggestion has definite merit, especially when samples from different locations are to be studied (Phillips, 1980).

This can be seen with the US Mussel Watch (Goldberg et al, 1975) where bivalves were sampled wherever possible, within a given size range of 5 to 8 cm. It should be noted that this range is still considerable, but the bulk of available evidence for organic contaminants suggests that most of the size-correlated effects are secondary to size-lipid correlations (Phillips 1978;1980). If lipids are quantified in the samples studied, the source of variance may be accounted for (Phillips and Segar, 1986). Although data on invertebrates is sparse studies of *Chlamys opercularis* (scallop) have revealed no weight dependence of organochlorine concentrations (Ernst et al, 1976).

5.5.4 Variation with Length

Significant increases in wet weight-based concentrations of both sigma DDT and dieldrin have been shown to occur with fish length in two freshwater fish from the Great Lakes (Reinert, 1979). This length-related increase in organochlorine concentrations is, however, much less on a lipid weight basis than on a wet weight basis. The correlation between lipid and organochlorine data suggests that basing sigma DDT concentrations on lipid weights should completely eliminate any age dependence or length dependence of the data. Other influences also affect this pattern since residual increases in sigma DDT levels with length have been shown even when concentrations are based on lipid weights (Reinert and Bergman, 1974). Nevertheless, it is clear that age-dependent variations in lipid contents of these fish are responsible for most, if not all, the changes observed in organochlorine concentrations with fish length.
The other important observation in these studies is that size-lipid relationships (and hence size-organochlorine relationships) may vary for a species between locations. This influence of location on the length-organochlorine concentration relationship has also been noted during the extensive monitoring studies of Frank et al (1974) in Ontario freshwater ecosystems. Length-concentration relationships were reported (using both wet and lipid weight basis for concentrations) for sigma DDT and dieldrin in 21 fish species. These authors also noted that length-concentration relationships were most marked amongst piscivorous fish, at least where dieldrin was concerned; in contrast, species feeding on benthic invertebrates or plankton exhibited weak or no dependence of dieldrin levels on fish length. Hattula et al (1978a), reported on the extensive studies of organochlorines in freshwater fish from a Finnish lake. The most extensive data reported by these authors concerned *Esox lucius* (pike), *Perca fluviatilis* (perch), and *Ambramis brama* (bream). *Esox lucius* and *Ambramis brama* both exhibited a direct dependence of lipid contents on fish length and sigma DDT. PCB concentrations were also length dependent as a result of the lipid changes. The observed liability of the size-concentration relationship obviously makes the elimination of the effects of sample ages or sizes in monitoring surveys extremely taxing.

Reports concerning the variation in organochlorine concentrations with age or size of marine or estuarine finfish are much less common than are those concerning freshwater species considered above. However, the data available (for example Zitko (1971) working on *Clupea harengus* (herring); Jensen et al (1972) working on both *Clupea harengus* (herring) and *Gadus morhua* (cod); Ten Berge and Hillebrand (1974) working on *Clupea harengus* (herring) and *Pleuronectes platessa* (plaice); Ernst et al (1976) working on *Tringla lucerna*(yellow gurnard)) suggest that saltwater species may also exhibit age dependence in organochlorine levels and, once again, these may often be attributed to the covariance of lipid levels of the fish with age. The changes in concentrations of DDT and its metabolites in *Pleuronectes platessa* (plaice) are, however, certainly not lipid dependent (Ten Berge and Hillebrand, 1974). Phillips (1980) suggests that they may be caused by changes in exposure levels as these fish migrate offshore.

5.5.5 Conclusions

In many cases, according to Phillips (1980), age dependence of organochlorines is caused partly or wholly by concomitant changes in the total lipid contents of organisms as they increase in age or size. The increases in size of the total lipid pool (as a percentage of tissue weight) in older animals affords a sequestration site for greater amounts of organochlorines.

In some cases, however, covariance of lipids is not sufficient to explain the effects of age on the accumulation of organochlorines. In these instances, factors other than lipids may be involved. For example, if organisms exhibit age dependent migration (as seen in many coastal finfish), the time-integrated exposure concentration of organochlorines may be expected to change.

The effects of age and size on contaminant levels in bioindicators have never been fully separated from each other, although there are theoretical considerations which suggest that they may differ both qualitatively and quantitatively (Phillips and Segar, 1986).

Whatever the mechanism is that produces age or size-related changes in organochlorine concentrations in a species, the implications for monitoring surveys are clear. If samples are biased for age or size, this parameter will interfere in inter-sample comparisons and may give rise to totally incorrect assumptions concerning the ambient availability of organochlorines in a study area. Whilst the use of lipid weights as a basis for organochlorine concentration may not be expected to entirely eradicate such an influence of organism age or size, in many cases the interfering effects will be at least partly reduced.

The inability of lipid weight-based comparisons to completely eradicate size dependence of organochlorine concentrations in some instances, necessitates care at sampling to ensure that organisms of any one species from all study locations are as similar as possible in terms of age or size.

5.6 Effects of Season and the Sexual Cycle

5.6.1 Introduction

Seasonality in the concentration of organochlorines and, therefore, possibly also other hydrophobic organic contaminants in biota is due to two major factors:

(i) seasonal differences in availability of organochlorines to aquatic organisms which may be correlated to pesticide application or industrial discharge rates etc;

(ii) seasonal fluctuations of lipid contents of organisms, most frequently associated with the sexual cycle.

Differences in seasonal profiles of organochlorines with location may be produced by either factor varying with location. The use of indicators where seasonal fluctuations in contaminants occur, therefore, should employ multiple sampling with time in order to compare concentrations of contaminants at the seasonal maxima at each different site.

5.6.2 Seasonal Variation in Contaminant Availability

Residue levels in higher organisms have frequently been found to exhibit seasonal profiles dominated by the episodic nature of organochlorine delivery. Such effects are more severe in freshwaters close to the site of pesticide application. Correlation of seasonal organochlorine profiles to the timing of upriver usage of pesticides was demonstrated in studies of freshwater mussels as indicators of DDT (and its metabolites), aldrin and methoxychlor in a Michigan river (Bedford et al, 1988). Many other studies have shown a correlation between seasonal changes in residue levels in biota, particularly amongst salmonoids, and seasonal changes in the application of the compounds (Johnson, 1968; Kelso et al, 1970; Reed 1966, Cole et al, 1967, and Sprague et al, 1971).

Organisms of coastal waters or estuaries receiving organochlorines from distant sites often exhibit seasonal fluctuations in residues which may correlate to both gross inputs and organism physiology. Butler (1969b, 1971, 1973) and Butler et al (1971) considered the seasonal profiles of organochlorines observed in bivalves used in the US National Monitoring Programme of pesticides in estuaries and found that these reflected almost entirely the availability of these compounds in run-off waters. They concluded that seasonal maxima of pesticides may occur during periods of low run-off since constant output of organochlorines from industry and sewage is less diluted during low rainfall periods. This is in contrast to agricultural catchments where organochlorine maxima are found at periods of high run-off since a greater amount of pollutant is flushed into estuaries under these conditions.

This is backed up by work by Gomme et al (1991), looking at the hydrology of pesticides in the Granta catchment, UK. The occurrence of pesticides in rainwater was found to be broadly consistent with times of local usage. In river water the number and concentration of pesticides is far greater during periods of higher flow. Monthly monitoring demonstrated a close relationship between detected pesticides and river flow which accentuates the differences in pesticide flux between periods of high and low flow.

Van den Broek (1979) could find no marked seasonal variations in chlorinated hydrocarbon levels which may have been connected with agricultural activities. However, significantly higher levels of organochlorine pesticides have been detected from an area bounded by extensive orchards (Wharfe and van den Broek, 1978). Several authors have produced results which indicate the presence of this simple relationship between organochlorine application for agricultural purposes and the seasonality of organochlorines.in biota of the surrounding aquatic environment see, for example, Hansen and Wilson (1970); Conte and Parker (1975) and Livingston et al (1978).

In some cases, the peak in organochlorines present in the water as a result of run-off subsequent to crop spraying may coincide with the lipid maximum in aquatic biota. Organisms frequently spawn in late spring or summer, when crops are most often treated with pesticides (Phillips, 1980). The simple profiles discussed above are always found in areas where the major organochlorine source is agricultural. If, however, the source is mainly industrial, as with, for example, PCBs, seasonal profiles of the residues in biota tend to be far more erratic (Gustafson, 1970). It is therefore important to consider the sources of contaminants in the planning of monitoring surveys (see Table 2.1a).

There may also be seasonal variations in contaminant availability due to competition with other organisms. This has been suggested as a reason for lower organochlorine levels in organisms in the summer season since there is competition for dissolved organochlorine with summer populations of phytoplankton and zooplankton, encouraged by the increased temperature (de Kock, 1983; Olsson, 1978).

Increases have also been observed due to increased uptake from sediments after sediment resuspension during rough winter weather in coastal areas. Care is needed when comparing seasonality of benthic and pelagic organisms.

Marked seasonal variations have been observed with organotin levels in coastal waters, and organotin peaks in *Crassostrea gigas* (Pacific oyster) can be correlated with these (Ebdon et al, 1988). Most sites in south west England showed similar trends, with highest concentrations being found in May at the beginning of the boating season coinciding with the launching of yachts. The levels then fall fairly rapidly before showing a late season peak which varied in time with each site. The latter was generally associated with repainting or hosing off activities for yachts. Up to 6 g TBT may be hosed from a single yacht painted with a co-polymer formulation (Waldock, 1986), however the organotin concentration was shown to fall away during the winter. Waldock et al (1987) reported that in 1986 in popular yachting centres, the TBT concentration in water ranged from <1 ng dm^{-3} in winter up to 1500 ng dm^{-3} in marinas in summer.

Two peaks have been demonstrated in the adult *Crassotrea gigas* (Pacific oyster) TBT concentration curve, which correspond to the peaks seen in the water concentrations (Ebdon et al, 1988).

TBT is associated with particulates in the water column and also sediment. TBT associated with both sources is bioavailable to oysters (Waldock and Thain, 1983). Sediment organotin concentrations have been found to be generally 1000 times greater than those found in the overlying water (Maguire et al, 1986; Valkins et al, 1986). Organotin residues in organisms closely reflect distribution of contaminants in waters and sediment (Ebdon et al, 1988; Langston et al, 1987). This has become less important since the use of paints containing TBT is now restricted, but breakdown products, DBT and MBT may remain in sediments for some time.

5.6.3 Effects of the Sexual Cycle

Several authors (Phillips, 1976, 1980; Romeril, 1979; Simpson, 1979; Zaroogian, 1980; Gault et al, 1983; Howard and Brown, 1983) have found that organism physiology has a marked effect on pollutant seasonality in biota. A change in lipid level in an organism may thus affect organochlorine flux markedly. Most organisms exhibit well-defined seasonal variations in lipid levels which correlate with the sexual cycle.

In general, aquatic organisms exhibit increases in whole-body lipid content during pre-spawning maturation. These changes in lipid content are due mainly to the enormously increased lipid levels of gonad tissues which contain large amounts of lipid-rich gametes. It is into these primary fats that the main partition of pollutants can occur.

In oysters, spawning leads to a loss of pesticides associated with the gametes. It is likely that the fall in PCB content observed in spring in mussels
Mytilus edulis (common mussel) (Hummel et al, 1990) and also in *Salmo gairdneri* (rainbow trout) (Guiney et al, 1979) is due to the same reason. In female teleosts, eggs may represent a high proportion of the total body load of compounds, while in the males the organochlorine loss in milt is relatively far less important, at least for *Salmo salar* (Atlantic salmon) (Anderson and Fenderson, 1970). The effects of lipid seasonality on organochlorine levels in biota have also been reported for many finfish (Anderson and Everhart, 1966), among crustaceans such as the *Callinectes sapidus* (blue crab) (Sheridan, 1975) and also in phytoplankton (Jensen et al, 1970).

Different species of bivalves taken in any one area can exhibit differences in the seasonal fluctuation of their accumulated organochlorine residues (Foehrenbach, 1972). There are two possible explanations for this phenomenon. First, bivalves may respond differently to the various exposure routes by which they can accumulate organochlorines. Secondly bivalves of different species spawn at different times of the year. If spawning (and the lipid cycle) is important in determining organochlorine concentrations, the seasonal profiles will be unique to each species.

Spawning effects may be overcome to a large extent by basing organochlorine concentrations on lipid weights but this assumes that water-lipid partitioning is the major uptake mechanism and that organisms are at equilibrium with their surroundings at all times. Hummel et al (1990) found that when PCB concentrations in *Mytilus edulis* (common mussel) are measured on a dry weight basis, in general levels increase during the winter, drop to low levels in spring before increasing again during the summer. However, on a fat basis, concentrations mostly showed a continuous decrease from autumn/winter to summer. Decreases in PCB levels coincided with an increase in fat content. It was found that *Mytilus edulis* (common mussel) may 'lose' half to two thirds of the PCB content by means of their reproductive output.

Other effects, for example different exposure routes, may play a role and are more difficult to make allowances for and thus a knowledge of local hydrology and input sources would seem essential in aiding correct interpretation. Olsson (1978) found that an additional factor that increased PCB uptake could be due to increased activity observed during the spawning season.

5.6.4 Effects of Migration

The seasonal fluctuation in trace organic residues in some organisms may be affected by migration. Many coastal teleost species, for example, spawn in estuarine areas and migrate to these grounds from feeding grounds further off shore prior to spawning. The seasonality of organochlorines in water is frequently more pronounced in estuaries than in offshore waters. Thus in some cases it has been suggested that migration was the basis of seasonal changes of, for example, DDE and dieldrin in *Gadus morhua* (cod), and *Ammodytes lanceloatus* (sand eel) from the Farne Islands (Robinson et al, 1967).

Migration was also put forward as an explanation for inconclusive seasonal results in the Medway Estuary (van den Broek, 1979) while earlier work by Wharf (1975) had found a summer rise in pesticides in biota associated with increased spraying activity. The study by van den Broek was at a more distant location from the pesticide source and since summer is a period of migration, it was suggested that many fish analyzed may have entered the sampling area only a relatively short period before they were caught. After analysing the seasonal variation of PCB levels in freshwater fish Olsson and Jenson (1978) concluded that any aquatic system should be monitored when it is most stable, and probably not during the reproduction period of the test species.

It is important to note that the migration of an animal species introduces problems of data interpretation if the animal is to be used an indicator organism (see Section 4.1). For example an unusually high concentration of a given pollutant in a pelagic teleost may reflect either acute exposure to the pollutant at a site close to its capture point, or a chronic exposure to the pollutant over a much larger area. The organic contaminant concentrations in migrating animals are at best a complex function of previous locations and exposure times. The prerequisite that an organism should be sedentary in order to be representative of the study area has been much abused by authors using indicator organisms to monitor pollution; for example, the use of pelagic fish is frequent. Phillips (1980) notes that "it is hoped that authors will pay more attention in future studies to the basic indicator requirements, at least if the intention is to complete meaningful monitoring studies".

5.6.5 Conclusions

Sampling once only at any single point in time is of very limited value, since differences between the timing of seasonal maxima and minima at different sites will elicit spurious conclusions concerning the relative abundance of trace organic compounds at the sites studied (Phillips, 1978). Multiple sampling with time is vital if interfering parameters are to be minimised.

The shape of the seasonal concentration profiles and/or the timing of seasonal maxima in concentrations may therefore depend on a number of factors, namely:

- the timing of the spawning period;
- migration;
- location studies;
- age or size;
- the use of wet or lipid weights as a basis for concentrations.

5.7 Effects of Interactions Between Contaminants

It is rare that only one stress occurs in the natural environment and more usually a number of different chemicals and interactions may occur whereby residues of one compound affect the uptake or retention of others. The existence of interactive effects in the aquatic environment between different trace organic compounds and other pollutants has been established for some organisms.

While the mutual effect of organochlorine pesticides appears to be either neutral or inhibitory (Ferguson and Bingham, 1966; Addison et al, 1976), there are exceptions. Work by Macek et al (1970) and Mayer et al (1970) showed that dieldrin appears to enhance the uptake and storage of DDT in *Salmo gairdneri* (rainbow trout). Both synergistic and antagonistic responses to mixtures of DDT and other organochlorines were noted for a marine diatom species (Mosser et al, 1974). Clarke (1986) reported that more than 50 of 209 PCB congeners are known or suspected to be inducers of mixed function oxygenase enzyme activities. As a result of field and laboratory work Shaw and Connell (1982) concluded that occurrence of petroleum hydrocarbons in sediments lowered the partition coefficient between water and sediments, decreasing the quantity of PCB available to polychaete worms.

The factors which may account for the ways in which different toxicants can influence the uptake of other xenobiotic substances have been summarised in EIFAC (1980) as follows:

- Detoxifying enzymes which can influence the uptake or loss of xenobiotic substances, may be inhibited.

- Permeability of membranes may be changed thereby affecting the rate of uptake of other substances.

- Internal physiological changes, such as increased rate of blood flow through the gills of fish, may increase the accumulation rates of other substances.

- There may also be external interactions of toxicants, perhaps in the formation of complexes which may influence uptake.

Interactions between contaminants should be considered because if the uptake of a given pollutant depends not only on its own ambient bio-availability but also on the presence, absence or precise concentration of a second contaminant then the bioindicator concept breaks down (Phillips, 1980). These effects are extremely important if they are likely to interfere with indicator surveys since no allowance can be made for them during either sampling of biota or in the interpretation of the results. Such effects have been reported for several fish species and this eliminates the possibility of using these species in the monitoring of organochlorines in the aquatic environment.

5.8 Effects of Environmental Variables

5.8.1 Introduction

Water quality variations which may take place with season can have a marked effect on the accumulation of organic contaminants by biota. Three changes in water quality may be considered as being particularly important:

- turbidity;
- salinity;
- temperature.

5.8.2 Turbidity

The importance of turbidity is allied to the tendency of trace organics and in particular organochlorines to adsorb onto inorganic particles. The exact importance of turbidity in the overall availability of organochlorines to aquatic biota is uncertain (Phillips, 1980). However, filter-feeders in the water column are likely to respond to residues adsorbed to particulates and many benthic organisms may also take up organochlorines from sediment particles for example Burnett (1971) found that the *Emerita analoga* (sand crab), contained increased concentrations of DDT after a winter period of stormy weather.

Other authors have also reported data suggesting that sediments may be a significant source of organochlorines particularly in marine and estuarine systems. For example Young et al (1976) working on *Mytilus californianus* (california mussel), and Fowler et al (1978b) working on PCB uptake in
Nereis diversicolor (common ragworm) suggested that perhaps as much as 90% of the body load of PCBs is sediment-derived. This predominance may be greater for benthic infauna than for epifauna because of the intimate contact of the former organisms with the sediment. However, this route cannot be discounted for any organism which lives in contact with the sea bed.

On the otherhand, Bedford et al (1973) found that if the suspended load was too great for *Unionidae* (freshwater mussels) they tended to reduce their filtering, and that very poor water quality could cause the mussel to close and not filter at all for a period of time. This would clearly affect uptake of organochlorines via this route.

5.8.3 Salinity

The effect of salinity on the accumulation of organochlorines has received very little attention to date, despite the fact that estuarine organisms are certainly at great risk from organochlorine contamination (Butler, 1969a ,1969b ,1971). It is possible that although absolute concentrations of contaminants in water are similar throughout a large area, estuarine biota within this area could nevertheless accumulate a greater or smaller residue load than open-sea biota because of some direct effect of salinity. If the intention of a monitoring survey using a biological indicator is to define areas of high trace organic contamination, then the inability to differentiate between absolute abundance and salinity dependent availability is a serious disadvantage.

Early work by Murphy (1970) found that a single salinity tolerant fish bioconcentrated DDT and related compounds to varying extents at different salinities. This has been reinforced by a number of studies illustrating that organochlorine content is inversely related to salinity (Davies and Dobbs, 1984; Duursma et al, 1986; Hummel et al, 1990; van Zoest and van Eck, 1990; 1991).
Goerke and Weber (1990) found that populations of *Nereis diversicolor* (common ragworm) eliminated PCBs at a faster rate in a mesohaline environment than in a polyhaline one. However, when populations were transferred and acclimated to control conditions, no difference was seen in the elimination rates. This suggests that the results were due to inherent properties of the population, such as genetic differences, or inducibility of enzyme systems (ego mixed function oxygenases). Others have suggested that the main way in which salinity acts upon bioaccumulation is through affecting uptake of contaminants Reduced uptake at high salinities may be due to a decreased organic carbon content and hence a lower number of sites for adsorption so that lower levels of organochlorines are held in the water

column (Davies and Dobbs, 1984). Decreased pollutant concentration may also be due to the mixing of riverine with marine particles (van Zoest and van Eck, 1990).

Degradation processes may also reduce the levels of pollutants at increased salinities, for example, volatile halogenated hydrocarbons. The decrease of these compounds cannot be explained by variabilities in riverine concentration, but it has been found that the volatization rate can be relatively high all year round, due to the relation between Henry's Law constant and temperature in the summer and high wind speeds in the winter (van Zoest and van Eck, 1991). The extent to which microbial degradation and photolysis also play a role depends upon the compound and the conditions.

Salinity was considered by Phillips (1978) to be an important factor in the uptake of organochlorines in estuarine species but the effects are still not fully understood. This is an important aspect for monitoring since it appears that organisms from different salinity conditions cannot be freely compared without this factor being taken into consideration. For example, the concentration of organochlorines on a fat basis has been found to be correlated with salinity but was only significant over a two year period. During shorter time spans, seasonal changes in fat content and individual variation reduced the significance of this relationship (Duursma et al, 1986). It seems therefore that whilst salinity effects are important they may be masked by other more variable factors.

5.8.4 Temperature

The effects of water temperature on the net uptake of trace organics by aquatic organisms have received rather more attention than have salinity effects. Temperature, for example, influences the chemical and physical state of pesticides (Muirhead-Thompson, 1971). Differences in temperature can affect the general activity, metabolism and behaviour of freshwater organisms in ways which can expose them, to a greater or lesser degree to pesticides in the water. The contaminant itself will probably be more soluble in water of higher temperature as noted for organochlorines (Biggar et al, 1966). As discussed by Phillips (1978), this effect of temperature will lead to a shift in the relative amounts of organochlorines in solution and in particulate-adsorbed forms.

Hamelink and Waybrant (1976) studied DDE and lindane kinetics in a natural freshwater ecosystem. It was noted that lindane remained mainly in solution throughout the experiment, whilst DDE was rapidly lost to the bottom sediments. It was later noted that DDE was significantly mobilised from the sediments during the summer; this effect probably reflects the shift in the equilibrium between adsorbed and soluble forms in response to warmer temperatures.

Temperature would be expected to affect the degradation of organic contaminants and the processes involved with this (de Koch, 1983). It also appears to influence the rate of uptake of toxicants, for example Veith et al (1979) found that several fish species showed increased bioconcentration factors of PCBs at higher temperatures and Reinert et al (1974) found a correlation between DDT uptake and temperature in *Salmo gairdneri* (rainbow trout).

Boryslawskyj et al (1978) found that the time taken to reach steady state concentration of dieldrin in the tissue of *Sphaerium corneum* (orb shell cockle) was inversely proportional to temperature; as the temperature increased, the time to equilibrium decreased and the rate of accumulation rose.

The toxicity of insecticides has been shown to be strongly affected by changing temperatures (Roberts, 1975c; Katz and Chadwick, 1961; Cope, 1965; Holden, 1972b). The responses have been divided into two main types (Ferrando et al, 1987):

- positive temperature coefficient - organophosphates, carbamate, cyclodienes, endosulfan.

- negative temperature coefficient - DDT, lindane and most pyrethroids.

Murphy and Murphy (1971) found that the relationship between accumulated DDT and oxygen consumption was almost linear. This may play a part in the greater toxicity observed with *Anguilla anguilla* (eel) (Ferrando et al, 1989), since at higher temperatures the organisms become stressed as a result of decreased dissolved oxygen content in the medium. They are then more vulnerable to the effects of pesticides since the stress causes an increase in the amount of pollutant ingested by the organism.

It seems that the mechanism by which the organism responds to the changes in water temperature is mediated by general metabolism. It acts in a variety of diverse, interacting ways, eliciting alterations in absorption, distribution, penetration of organs, detoxification, excretion and differential target site interactions (Canyurt, 1983). It can also affect feeding rates, filtration or pumping rates. For example Boryslawskyj et al (1987) demonstrated that the rate of accumulation of dieldrin in a freshwater bivalve increased with temperature, which was related to the frequency of gill cilia beating, altering the volume of water extracted.

The effect of temperature on the uptake of, for example, organochlorines probably also contributes to the seasonality of these compounds in biota. These effects are difficult to separate, however, from those of variations in the available ambient concentration of organochlorines and lipid changes (Phillips, 1980). Alterations in the lipid chemistry of aquatic organisms will have marked effects on the ability to sequester organochlorines in their tissues.

Ambient temperature is an extremely important determining parameter, because of its effect on the metabolic rates of organisms and also on the cycling of organochlorines in the environment (Phillips 1978). However, it appears that no generalisations can be made concerning the effects of temperature on the toxicities of organochlorines as differences certainly exist between different compounds and different species.

It is evident that any study of trace organic concentrations in biota during an indicator study, irrespective of the size of the study area, should take water temperature into account as a probable influencing factor.

5.8.5 Conclusions

It is clear from this discussion that the changes in water quality which may occur with season can have a direct bearing on the accumulation of organic contaminants by biota. This is true whether dealing with a lake, river, estuary or coastal situation. A typical case study which encapsulates this is, for example, a temperate estuarine ecosystem which might exhibit a winter season of high run-off causing turbid waters of low salinity to flood the estuary; both these effects would tend to increase contaminant uptake by biota, although the low winter temperatures would tend to have the opposite effect. It should be emphasised however, that in most cases the effects of these parameters will be secondary to the more important changes which occur with season in contaminant availability in the environment and the lipid content of aquatic organisms.

5.9 Effects of Contaminant Toxicity

Although field and laboratory studies have not been extensive, contaminated sediments have been shown to lead to increased mortality, decreased growth and fecundity, and morphological anomalies (Knezovich et al, 1987). If chemical toxicants are present they may pose a long term threat to the aquatic ecosystem.

The time scale of the accumulation of the toxicants can also play a role. While *Crassostrea virginica* (eastern oyster) will tolerate tissue residues of DDT at least as high as 150 ppm without apparent ill effect, this only occurs if the residues are accumulated slowly. However as little as 0.1 ppm DDT in the surrounding water can terminate feeding activities in *C. virginica* and at summer water temperatures (31°C) will cause death (Butler, 1973).

The body weight of fish affects their rate of organochlorine uptake from food. Buhler et al (1969) and Buhler and Shanks (1970) have found that the toxicity of dietary DDT is greater to smaller individuals.

Some studies of pesticide toxicity have also shown that while many are less toxic in hard water, others are not influenced by hardness and some are more toxic in softer water (Holden 1973).

Reproduction is often considered a susceptible stage of the life-cycle and fecundity is affected by contaminants, for example in *Capitella capitata* (Chapman and Fink, 1984), in fish (Mayer et al, 1970), in daphnia (Canton et al, 1975), and in *Nucella lapillus* (common dogwhelk) (Bryan et al 1986). Some pesticides induce an increase in the breakdown of certain steroids, for example sex hormones which could cause inhibition of reproduction (Peakall, 1967; Kupfer, 1967). While the mechanism underlying imposex in *Nucella lapillus* (common dogwhelk) produced by TBT is not clear it is believed that it may also involve an induced hormonal imbalance (Smith, 1981c; Feral and Gall, 1982).

Other effects have been seen with infaunal polychaetes, bivalves and amphipods which showed impaired burrowing behaviour when placed in pesticide contaminated sediment

(Gannon and Beeton, 1971; Mohlenberg and Kiorboe, 1983). Streit and Peter (1978) demonstrated that under experimental concentrations of 16 mg l^{-1} atrazine *Ancylus fluviatis* (river limpet), could not attach to the walls and so would die quicker in a natural habitat.

The existence of such toxic effects in the environment is extremely difficult to elucidate unless researched in some detail. Several authors have, however, shown that populations can clearly be affected by organochlorine toxicity and that this will influence their use in certain situations as a bioindicator.

5.10 Trophic Level

The evidence for contaminant 'biomagnification' through food chains in aquatic environments is conflicting (Johnson, 1973; Phillips, 1980). Not all substances are accumulated in food. Some compounds reach similar concentrations at all trophic levels and in addition, when differences do occur they cannot always be explained solely by position in the trophic chain. Consequently authors for example Portmann (1975) and Hargrave and Phillips (1976) discussing this subject have reached opposite conclusions according to their selection of examples from the literature.

Woodwell (1967) observed the stepwise increase of contaminant concentration with increasing trophic level and cited it as evidence of biomagnification. Macek and Korn (1970) consider food-chain concentration to be the major source of contaminants. Van den Broek (1979) provides some evidence of increases in organochlorines along food chains, reporting greater levels of contaminant found in the gut wall than in the musculature of *Sprattus sprattus* (sprat). He was also able to shown higher concentrations in *Pomatoschistus minutus* (sand goby) than in *Crangon vulgaris* (brown shrimp), the former being at a position further along the food chain than the latter. Work conducted by van der Oost et al (1988) suggested that biomagnification contributed significantly to the total PCB concentration and that this was most pronounced for higher trophic food chain organisms. It was reported that TBT accumulation via *Artemia nauplii* (brine shrimp) consumed by adult *Rhithropanopeus harrisii* (mud crab) was the dominant uptake route rather than uptake directly from water (Evans and Laughlin, 1984). Robinson et al (1967) reported that residues of organochlorine insecticides tend to be greater in marine organisms of higher trophic levels, but this tendency was not found in all food chains. Biomagnification may occur but it probably does not result in major increases in pollutant concentration (Connell, 1988). Bryan (1979) concluded that although food is often the most important source of pollutants for marine species, it does not always follow that predators at higher trophic levels contain the highest pollutant concentration. There are many cases where food chains do not exhibit this phenomenon (Shaw and Connell, 1982; Reinert, 1972; Laughlin et al, 1986).

A number of papers describe experimental work utilizing feeding experiments of various kinds to evaluate the contribution of water and food to contaminant loads in selected organisms. Experiments by Reinert (1972) with dieldrin showed that daphnia and guppies accumulated more from water than from food when they were exposed to similar concentrations in water. Canton et al (1975) investigated the *Chlorella-Daphnia-Lebistes* food chain transfer of HCH and concluded that transfer from water was the most important, but that food was a significant source. In studies of the algae-daphnid and daphnid-catfish system Ellegenhausen et al (1980) determined that water was the more significant source of pesticide residues. Within invertebrate food chains both Kerr and Vas (1973) and Rosenberg (1975) concluded that there was no clear evidence for accumulation with trophic level.

It is difficult to interpret all these results and to arrive at an overall conclusion. Hellawell (1986) suggests that factors such as longevity, size, lipid content and the excretion efficiencies of organisms, some of which also correlate with trophic level, could equally be responsible for the apparent differences. Connell (1988) offers additional parameters such as feeding rates, metabolic rates, physical form of food and other chemical components in the food that may also play a role. Biomagnification may be explained, not in terms of increasing concentrations through predation but by the lipid contents of various organisms. The pollutants partition between water and lipid resulting in increased residues in organisms at higher trophic levels simply because of the greater fat content (Hamelink et al, 1971; Hamelink and Waybrant, 1976; Portmann, 1975). Most of the data regarding trophic level variation in organochlorines suggest that organisms absorb these pollutants more rapidly from water than from food, and that the contribution, per unit time, of residues from the food chain to the total body load of an organism is rather less than 30% (Phillips, 1980).

An important parameter which defines the relative uptake of organochlorines from each route is the relative concentration of these compounds in the food and water to which the organism is exposed. Thus Macek and Korn (1970), using a ratio of 10^6 for DDT (3 μg/g in food and 3 ng/l in water) found predominance of the food route, whereas other studies (Reinert, 1967; 1972; Lenon, 1968; Chadwick and Brocksen, 1969; Jarvinen et al, 1977), all of which used food: water exposure concentration ratios between one and two orders of magnitude less than that of Macek and Korn, argued that uptake of organochlorines from solution was the more important route.

While both bioconcentration and biomagnification vary in importance with organisms in various groups depending on a variety of factors, in general the former is of greater significance. Phillips (1980) suggests that lipid-water partitioning is more important to organisms of lower trophic levels, whilst the uptake of organochlorines from food is predominant in biota of higher trophic levels. The result of this is a general increase in the concentration of organochlorines on a wet weight basis with increased trophic level; lipid weight-based data would exhibit a less marked increase. Portmann (1975) showed that with DDT concentrations calculated on a lipid rather than a wet mass basis the magnification of concentrations along the food chain phytoplankton-seals was reduced from several orders of magnitude to less than one order.

5.11 Conclusions on the Factors Affecting Indicator Organism Reliability

The major factor determining the concentrations of organochlorines in biota (other than the ambient levels in their surroundings) is the amount of lipid present in the organisms studied.

The use of a lipid weight basis for contaminant concentration eliminates the differential effects of lipids but this is still based upon certain assumptions.
The use of wet weights as a basis for organochlorine concentrations in indicator surveys is suspect if the intention of the survey is to produce meaningful data on the relative abundance or biological availability of organochlorine pollutants at different locations (Phillips,1978). The situation is complicated, however, since there are many interwoven factors that have an effect upon the amount of lipid present in an organism and hence upon the concentration of contaminants in biota. These factors affect the reliability of an indicator organism and it has been recommended that the concentration of contaminants in survey biota should be reported using both wet weight and lipid weight values. Lipid content of an organism varies both with age (and size) and also sex, and hence to account for these variables organisms which represent a range of physical states should be taken for each species studied at each location. As the lipid content increases, so the capacity for accumulation of contaminants increases since there is a greater volume of lipid for them to partition into.

Phillips (1978) recommends that each location should be sampled on several occasions to compensate for the non-simultaneous timing of seasonal maxima and minima in organochlorine concentrations in organisms of the same species from different locations. This is important since variations in both availability of contaminants due to application and discharges, and also physiological state of the organism vary with season. Seasonality of the physical environment in terms of contaminant availability, turbity, salinity and temperature play a role in the bioavailibility of trace organic compounds but while these effects are important it is likely that they will be overshadowed by the effects of variations in lipid content. If bioaccumulation is to be used as a valuable technique for monitoring trace organic compounds these factors need to be taken into account in the careful design of a monitoring programme.

6 CURRENT RESEARCH AND USE OF INDICATOR ORGANISMS

6.1 Introduction

In the proceeding sections the emphasis has been concentrated on providing a sequential understanding of biaccumulation as an approach for monitoring Red List trace organics, looking at its potential for the compounds of interest, the mechanisms of the process, the advantages of the approach and the factors affecting the use and reliability of organisms.

In this section the use to date of different groups of organisms in both freshwaters and estuarine/coastal situations is discussed. There is inevitably some overlap with preceding sections since current use of certain organisms has necessitated an in depth look at factors affecting bioaccumulation.

The organisms considered under the two main headings of fresh and estuarine waters have been put into three major groups - plants, invertebrates and fish.

6.2 Freshwater Organisms

6.2.1 Plants

Algae

It seems that algae in general have been largely overlooked as suitable bioaccumulation monitors of trace organic compounds in freshwaters. This may in part relate to the previous focus on organochlorine compounds and their affinity for lipids. Since higher amounts of lipid were generally associated with aquatic animals this would tend to make them a natural choice for bioaccumulation studies of organochlorines leaving plants and in particular algae largely ignored. In a review of the use of algae for monitoring rivers, Whitton (1991) suggests that several macro-algae could be potentially very suitable as monitors of trace organics. He does however point out that nutrient regime, seasonal changes (in nutrients, light, temperature etc) and other environmental factors all affect cell composition and hence lipid content in algae. This means that considerable research is needed before algal tissue analysis can provide a quantitative approach to monitoring trace organics in rivers.

To date there are only scattered reports on the accumulation of trace organics by freshwater algae and apparently no accounts on their applied use in biomonitoring programmes. *Chlorella*, a unicellular green alga, has been found to have a great capacity for accumulating DDT (Södergren, 1968) and dieldrin (Wheeler, 1970). However the usefulness of this genus as an indicator is questionable considering its microscopic size and difficulty in identification.

It has been suggested that *Cladophora* (blanket weed) would serve as a suitable indicator organism for DDT since it has been demonstrated that it is especially efficient in concentrating this organochlorine compound (Meeks and Peterle, 1967; Woodwell, 1967; Ware et al, 1968).

It has also been successfully used as a bioindicator for PCBs in a wide ranging monitoring programme along the Niagra River in Canada (Niagra Rivers Toxics Committee, 1984). Since the earlier studies on DDT there appears to have been very few additional investigations on the use of *Cladophora* for detecting and monitoring other trace organic compounds. This does appear to be surprising since for the bioaccumulation of metals there has been comparatively much more work done (Whitton et al, 1989; Whitton, 1991; Whitton et al, 1991). *Cladophora* would certainly seem to merit further investigations since some of the advantages identified for its use as a monitor of heavy metals (such as the rapid response of internal concentration to external concentration in the water as compared with other organisms) might similarly apply to trace organic contaminants.

Mosses

Until comparatively recently mosses too were largely ignored as potential bioaccumulation monitors for trace organic compounds. However following a considerable amount of research on their use as monitors of heavy metals in both the UK (Say et al, 1981; Say and Whitton, 1983; Wehr and Whitton, 1983; Wehr et al 1983; Whitton et al, 1991) and in Belgium and France (Empain, 1974; Empain et al, 1980; Mouvet, 1978; Mouvet, 1985; Mouvet et al, 1987) studies by these authors have extended the research on metals to include investigations into the potential use of aquatic mosses as biomonitors of trace organic compounds.

Research has shown for example that aquatic mosses can accumulate PCBs and gamma-HCH (Frisque et al, 1983), and that *Fontinalis* (willow moss) accumulates the organochlorines lindane and dieldrin under laboratory and field conditions (NECL,1989). Mouvet et al (1985) demonstrated that the aquatic moss *Cinclidotus danubicus* can be used to locate sources of organochlorine pollution. The observed concentrations were greater or equal to values obtained for sediments, plankton, other hydrophytes, invertebrates or fish. Mouvet et al (1985) summarised the advantages of mosses as follows:

- Sedentary behaviour of aquatic mosses makes hand sampling easy and enables the precise location of the pollutant to be found.

- Their morphology minimizes the problems due to the differentiated organs that are present in fish and aquatic macrophytes.

- Tolerance to various types of pollutants enables their use for a wide range of studies.

- Accumulation factors are quite high for example 234 for gamma- HCH and 4865 for PCBs.

Recent work (Gallissot and Mouvet, in prep) shows that native mosses from very polluted water can contain 68 $\mu g\ g^{-1}$ HCH and 10 $\mu g\ g^{-1}$ PCBs. The characteristics of mosses that explain these high levels include the following:

- Tolerance of a very high level of water pollution (0.7 $\mu g\ gl^{-1}$, 10 $\mu g\ gl^{-1}$ PCBs, Gallissot and Mouvet, in prep).

- Being attached to boulders and other permanent substratum, they are permanently exposed to pollutants and can not seek refuge in less polluted areas.

- Life span of more than one year allows a long exposure time.

- The surface in contact with the environment is very large since the leaves are essentially unilayered.

- They have no excretion system like invertebrates and fish.

- Their lipid content (1.4-4.0 %, Frisque et al, 1983) is quite high.

The the ease of transplanting aquatic mosses, as demonstrated by Mouvet et al (1985) is a great advantage as compared with other biological elements of the aquatic ecosystem for the following reasons:

- It facilitates comparison of various geographical locations by employing statistically similar groups of plants derived from a common stock.

- The period of exposure to the polluted environment is known.

- The investigator can select monitoring locations independent of the natural occurrence of the indicator species.

Recent research on the effect of herbicides (in this case atrazine) on submersed macrophytes at concentrations typically encountered in the environment, has shown that not only does the aquatic moss *Fontinalis antipyretica* (willow moss) tolerate these levels but also takes up and binds the herbicide (Hofmann and Winkler, 1990). *Fontinalis antipyretica* (willow moss) has also been shown to accumulate atrazine and simazine under laboratory conditions (NECL, 1989). However, recovery of atrazine was found to be low (30-40%) and this was thought to be related to the high water solubility of the compound.

6.2.2 Freshwater Invertebrates

Leeches

Freshwater leeches have been shown to bioaccumulate organic contaminants, including lindane, DDT and PCP, to levels exceeding those reported for other aquatic organisms (Metcalfe et al, 1988). The contaminants were bioaccumulated in proportion to their relative occurrence in water but no clear pattern relating the degree of chlorination and chemical properties of various isomers to their bioaccumulation potential in leeches was evident. While lindane was rapidly eliminated from leeches, with a half-life of a day or less, chlorophenols and DDT derivatives are only slowly depurated. In most aquatic organisms, the biological half lives of chlorophenols are in the order of a few hours to a few days, but Metcalfe et al (1988) demonstrated that half lives for these compounds in leeches were in most cases longer than one month. The only other organisms able to accumulate chlorophenol residues to a comparable magnitude to those found in leeches, as demonstrated by these authors, were oligochaete worms, which belong to a class of annelids that are closely related to leeches. This information suggests that annelids in general may have unusually high bioconcentration capacities for chlorophenols.

Webster (1967) reported the presence of DDT in leech tissues (*Erpobdella punctata*) three months after the aerial spraying of a marsh, while no residues were found in amphipods or copepods. Meeks (1968) found that DDT residues in an experimentally sprayed marsh were higher in the leech *Erpobdella punctata* than those in molluscs, crustaceans and insects.

Leeches are promising candidates for biomonitoring of pollutants since they are able to accumulate DDT, Mirex and chlorophenols without being killed and since they are relatively sedentary they are representative of the study area (Metcalfe et al, 1984). Leeches have been found to be abundant in littoral zones in streams, lakes and ponds, and both in pristine and polluted waters. They are of a reasonable size, yielding adequate tissue for analysis, ideal for laboratory experiments easy to sample, and Metcalfe et al (1984) concluded that erpobdellid leeches are potentially the most suitable for biomonitoring purposes. Several studies have reported, however, narrower tolerance ranges for both temperature (Linton et al, 1983a) and dissolved salts (Reynoldson and Davies, 1980) in post-reproductive leeches. Metcalfe and Hayton (1989) make the following recommendations for the design of surveys using leeches as bioindicators:

- Specimens in reproductive condition should be avoided as they tend to be in poor condition, with lower resistance and since they are nearing the end of their life cycle, natural mortality rates will be high.

- Studies should be conducted at a time of year when water temperatures are cooler, and therefore more favourable.

The pioneering work proposing leeches as indicator organisms was carried out by a group with the National Water Research Institute, Canada and no published literature on the use of leeches in the UK was discovered during the present review.

Other

The use of other freshwater invertebrates as bioaccumulators of Red List trace organics has received comparatively little attention. The following case studies, however, may be mentioned to illustrate the potential use of these organisms in freshwater ecosystems:

- *Gammarus pulex* (freshwater shrimp) has been successfully used by Sodergren et al (1972) as an indicator of chlorinated hydrocarbon residue distribution in southern Swedish streams. These workers also demonstrated that stoneflies (specifically *Brachptera risi, Capnia bifron, C. nigra)* and *Ephemera danica* (mayfly) were good accumulators of chlorinated hydrocarbons.

- *Sphaerium corneum* (orb shell cockle), a freshwater bivalve, has also been successfully used as an indicator of dieldrin in freshwaters in West Yorkshire (Boryslawskyj et al, 1987).

- PCB uptake has been demonstrated in particle-feeding chironomid larvae (Larsson, 1984).

- Mauck et al (1976) found that simazine residues in aquatic fauna can last in excess of a year, with bioaccumulation occurring in invertebrates.

6.2.3 Freshwater Fish

In Britain comparatively few data on residue levels in freshwater fish have been published. Hider et al (1982) conducted a wide ranging study of chlorinated hydrocarbons in freshwater fish between 1980 and 1981. At a detection level of 0.01 mg kg^{-1} only 9.7% of individual fish were contaminated with pesticides with DDE being the most frequently detected. PCBs were not detected. Significantly more *Anguilla anguilla* (eel) than other species contained measurable amounts of pesticides, (see below) and this is probably related to the high lipid content of the muscle tissue of this species.

Holden (1973) found the following levels in *Anguilla anguilla* (eel):

DDT	0.36-0.8	mg kg^{-1}
Dieldrin	0.52-0.95	mg kg^{-1}
PCB	<0.1	mg kg^{-1}

Leuciscus rutilus (roach) has been used by Olsson et al (1978) as a monitor of PCB levels in Sweden. *Esox lucius* (pike), because of its relative stationary habit ie. restricted home range, was chosen by Olsson and Reutergardh (1986) as an indicator species in large Swedish lakes.

Hider et al (1982) conclude that with the withdrawal from general use of persistent pesticides within the UK, residues in fish have generally tended to decline.

Accumulation of TBT in fish has been demonstrated for farmed Atlantic salmon (Davies and McKie, 1987).

Eels

The life cycle of the eel is unique among European fish. During this cycle it experiences both marine and freshwater habitats, and especially in the latter stage it is exposed to a variety of anthropogenic substances. Work by Hider
et al (1982) found that eels were significantly more contaminated than other freshwater fish species, and these observations support previous suggestions that eels may be important biomagnifiers of chlorinated hydrocarbons (Holden, 1973).

Eels have been found to accumulate aldrin and dieldrin levels to concentrations exceeding those found in other fish, such as *Salmo salar* (Atlantic salmon),
Salmo trutta (trout) and *Phoxinus phoxinus* (minnow) (Hamilton, 1985).
Anguilla anguilla was found to be more contaminated by the organochlorines DDT, DDD, lindane and dieldrin than were marine fish such as *Gadus morhua* (cod) and *Clupea harengus* (herring) (Huschenbeth, 1977). Larsson (1984) stated that eels that live and feed in direct contact with sediment may be exposed to higher amounts of PCB than fish in the open sea. While PCBs are taken up directly from the water the highest concentration (471.6 $\mu g\ g^{-1}$) was found when eels were allowed to feed on benthic macroinvertebrates (chironomids) in sediment as a result of accumulation through the food chain.

The eel is generally thought to be a resistant species owing to its apparent robustness in the face of environmental fluctuations and its high tolerance to poor water quality, such as low oxygen content and high amounts of anthropogenic pollution (Tesch, 1977; Kruse et al, 1983; Ferrando et al, 1987). In addition they may survive for several years without food (Boëtius and Boëtius, 1985) and may inhabit waters influenced by high industrial activity, such as the River Elbe (Kruse et al, 1983).

There have been a number of eels killed, however, due to toxic effluents and it is now established that eels are more susceptible to toxicants than has generally been assumed. Several authors (Eister, 1970; Grauby et al, 1973; Aubert et al, 1977; Marchand, 1981; Canyurt, 1983; Ryan et al, 1984; Hamilton, 1985; Bertrand et al, 1986) agree with the opinion that the eel is more vulnerable than other common fish species, molluscs and crustaceans inhabiting contaminated fresh and marine waters. Despite this eels have been considered as suitable organisms for bioaccumulation studies for a number of reasons namely:

- They contain large amounts of fat, up to 40% of total body weight (fresh weight) which is stored mainly in muscle, and to a lesser extent in the liver and around the viscera (Henderson and Tocher, 1987). This is far higher than other freshwater fish, for example *Esox lucius* (pike) contains less than 2% fat in the muscle, which is a typical value for many North European freshwater species.

- The muscular fat deposits in eels serve as energy reserves and act as a sink for hydrophobic pollutants (see Section 2.4.2), accumulating organic contaminants to high levels.

- An important feature can be found in the reproduction of the eel. Anguillid eels, unlike many fish, spawn only once in their life, after migration to the Sargasso Sea. Hence the effects of the sexual cycle discussed in section 5.6 can be excluded and other relationships, such as age-pollutant concentration relationship can be more easily determined.

- Eels are long-lived organisms, allowing the sampling of more than one year class if required.

The age of eels has been significantly correlated to weight and length, and it has been found that the concentration of PCB and DDT and its metabolites increases with age. The largest increase in PCB and DDT uptake occurred in eels of 12 years or over. By this age the growth rate is stabilised and the dilution of pollutants by the growing biomass in younger year classes is no longer observed, leading to greater increases in organic contaminant concentrations per unit weight. The fat content also increases with age and weight (Lovern, 1938), and there is an inverse relationship between fat content and pollutant levels . While these correlations between age and pollutant concentration are significant on a fat weight basis, they are more so, when based on fresh weight. Increases in organic contaminants in older fish, however, may also be affected by a shift in diet with age (Larsson et al, 1991). While young eels consume mostly zoobenthos, as they grow, fish are a larger portion of their diet (Tesch, 1977) which may in turn contain higher pollutant levels.

Lindane concentrations in eels showed an inverse relationship with age. It has been shown that lindane is metabolised and eliminated to a higher extent than DDT in fish (Murty, 1986), and so it is probable that the efficiency of the metabolic processes generally increase with age (Rand and Petrocelli, 1988) as detoxifying enzyme systems develop and are induced from juvenile to adult fish (Larsson et al, 1991).

Recent decades have seen diminishing catches in Europe and a number of suggestions have been put forward to explain this, such as osmoregulatory stress and impaired gas exchange. It has been shown that organic contaminants for example DDT and PCB can disrupt osmoregulation (Kinter, 1972; Dave et al, 1975) and also can cause non specific changes to gill tissue that correspond to defense responses to any stressor agent (Miossec and Bocquene, 1986). Changes in predator-prey relationships have also been considered as a main cause for mortality and it has been suggested that eels avoid waters with a low pH (<5.5) (Brusle, 1991). In addition to the muscular fat deposits which contain much of the pollutant load are energy reserves for the long migration to the spawning grounds. As the deposits are depleted, the lipophilic pollutants are released into the bloodstream and can be transferred to vital organs and germinal tissue. Although reproduction potential seemed weakened by pollutants (Lopez et al, 1981a,b) scientific proof is lacking. While Brusle (1990b) claims that the decrease in the recruitment of elvers throughout European coastal waters is of anthropogenic origin he concludes that no clear-cut cause could be identified and further data were required.

6.3 Estuarine/Coastal Organisms

6.3.1 Plants

Estuarine Algae

As with freshwater algae, estuarine and marine algae have largely been ignored with reference to monitoring trace organic pollutants. It would seem that again preference has been given to macroinvertebrates and fish for obvious reasons including their higher lipid content, important presence in food webs often leading to man, and their commercial importance.

As was the case for freshwater algae, marine and estuarine species have been widely studied for their accumulation capacity for heavy metals (see for example the work of Bryan (1969); Barnett and Ashcroft, (1985); Say et al, (1990)). However in comparison very little work has been done on trace organic contaminants and the little that has appeared to be restricted to a few isolated accounts of accumulation.

For example Parker and Wilson (1975) demonstrated the use of macroalgae in the monitoring of PCBs in the Clyde estuary. *Pelvetia* and *Fucus* (species not given) appeared to accumulate higher PCB levels than *Ascophyllum nodosum* (egg or knotted wrack). The results showed similar trends to the PCB residue values quoted for mussels (Waddington and Mackay, 1972).

Metabolism of TBT has been reported for the green freshwater alga *Ankistrodesmus falactus* (Maguire et al, 1984). Langston et al (1987) suggest that a similar process may account for the limited accumulation of organotin and poor indicator properties observed in the seaweed *Fucus*.

Various species of marine algae accumulate dieldrin to levels many times higher than the original concentration in the medium (Rice and Sikka, 1973), but the degree of concentration of dieldrin by these algae was considerably less than that observed with DDT. This may be explained by differences in the water solubility of the two pesticides and hence the differences in their affinities for cellular lipids and uptake by algae.

6.3.2 Estuarine Invertebrates

Marine invertebrates have long been established as bioaccumulation monitors of trace organics in marine and estuarine systems particularly with reference to pesticides and PCBs (see Butler, 1973; Butler et al, 1971; Farrington et al, 1984; Eisenberg and Topping, 1984; Marcus and Renfrow, 1990). A number of these studies have illustrated the differing sources of organics either through absorption directly from the water, via suspended particulates during filter feeding or via contact with contaminated sediments.

PCBs in particular have been widely studied in sediments and it is found that they are taken up by benthic macroinvertebrates from contaminated sediment (Nimmo et al, 1971; Courtney and Langston, 1978; Fowler et al, 1978; McLeese et al, 1980; Sodergren and Larsson, 1982). The uptake is governed by processes such as ingestion of contaminated sediment particles (Nimmo et al, 1971), absorption of PCBs from interstitial water (Mcleese et al, 1980) absorption of PCBs leached to the water (Nimmo et al, 1971) and the uptake/exchange of PCBs directly from the sediment particles (Omann and Lakowicz, 1981; Larsson, 1983).

A number of groups of macroinvertebrates have been used over the years for monitoring and those considered for use by the Ministry of Agriculture, Fisheries and Food Directorate of Fisheries Research are given in Table 6.3a. Some of the more important ones are now dealt with in turn.

Table 6.3 a ***MAFF Directorate of Fisheries Research Shellfish Species Studied for Bioaccumulation***

Shellfish	
Abra	*Abra alba*
Crab - edible	*Cancer pagurus*
Cockle	*Cardium edule*
Crawfish (spiny lobster)	*Palinurus elephas*
Deep-water prawn	*Panalus borealis*
Faroe sunset shell	*Gari fervensis*
Hermit crab	*Eupagurus bernhardus* *Eupagurus prideauxi*
Lobsters	
Common	*Homarus gammarus*
Norwegian	*Nephrops norvegicus*
Mussel	*Mytilus edulis*
Oysters	
Japanese (Pacific)	*Crassostrea gigas*
Native Flat	*Ostrea edulis*
Pelican's foot shell	*Aporrhais pes-pelecani*
Queen (queen scallop)	*Chlamys opercualis*
Razorshell	*Cutellus pellucidus*
Scallop	*Pectem maximus*
Sea mouse	*Aphrodite aculeata*
Shrimps	
Brown	*Crangon crangon*
Burrowing	*Calocaris macandreae*
Pink	*Pandalus montagui*
Offshore brown	*Crangon allmani*
Smooth Artemis	*Dosina lupinus*
Starfish	*Asterian rubens*
Striped Venus	*Venus striatula*
Tubeworm	*Pectinaria koreni*
Towershell	*Turritella communis*
Whelk	*Buccinum undatum*
Winkle	*Littorina littorea*

Mussels

The use of mussels as bioindicators was first suggested by Goldberg (1975) and they have since been extensively used as quantitative bioindicators of marine pollution. The filterfeeding genus *Mytilus* appears to fulfil the basic prerequisites of a monitoring organism as listed by Phillips (1977, 1980); wide distribution, abundance, size, ease of collection, long life-span, euryhalinity, accumulation capacity and high concentration factors for several xenobiotics. Mussels have been recommended by many as the most suitable organisms for bioaccumulation of trace contaminants from marine or estuarine waters (Phillips, 1980). They have been used in internationally known monitoring schemes, notably the Mussel Watch concept (Goldberg et al, 1978; National Academy of Sciences, 1980).

Mussels have been used to detect PCBs in British coastal waters (Waddington and Mackay, 1972; Holdgate, 1971) and Cowan (1981) has successfully used *Mytilus edulis* (common mussel) to determine the extent and range of pollution of Scottish coastal waters by pesticides and PCBs.

This species is one of the seven "core" species used for metal, organochlorine pesticide and PCB monitoring by the Ministry of Agriculture, Fisheries and Food Directorate of Fisheries Research (MAFF, 1990).

Mussels have been studied to identify factors that affect bioaccumulation. While they are essentially isolated from uptake of organic contaminants from sediments by direct contact due to their hard shell, mussels are excellent for the study of filter and deposit feeding pathways (Knezovich et al, 1986). The kinetics of accumulation and elimination have been investigated in a number of studies.

Since large volumes of water are filtered by mussels and bivalves it suggests that the solubility of different PCB components would influence the ability of these animals to excrete different chlorinated biphenyls (Calambokidis et al, 1979). These results were backed up by those of Geyer et al (1982) who reported an obvious linear inverse relationship between log water solubility and log bioaccumulation of organic chemicals by *Mytilus edulis.*

Tanabe et al (1987) have also demonstrated that the biological half lives of individual PCBs in *Perna viridis* (green-lipped mussel) are generally very short, all being less than twelve days. This is obviously of great significance to the design of monitoring surveys if such short half-lives exist in other bivalve species. With total tin and organic tin the half-lives in *Mytilus edulis* were somewhat longer, being 25 days and 40 days respectively.
Cowan (1981) further suggests that the time and usefulness of a flushing procedure requires further investigation since the half-lives of alpha- and gamma-HCH, dieldrin and DDD determined by Ernst (1977) imply that a statistically significant loss of contaminant would occur during a 24 hour flushing period.

Several authors have shown distinct seasonal variations in organic contaminant burdens in mussels (Hummel et al, 1990; Phillips, 1985; de Koch, 1983; Goldberg 1986). The seasonal cycle profoundly affects biochemical, cellular and physiological processes (Bayne, 1989), and Hummel et al (1990) reported that mussels may "lose" half to two thirds of their PCB content by means of their reproductive output. In addition the lipid content of *Perna viridis* (green lipped mussel) varied almost three fold which suggests that within the study of Phillips (1985) there was considerable variability in the sexual condition of the samples between locations.

Goldberg (1986) reports a number of reasons for the seasonal variation in concentrations seen in the Mussel Watch such as changes in the biological or biochemical activity of organism such as filtration rates or spawning. There may be changes in biological or biochemical activity of other organisms commonly associated with bivalves, such as microbial activity or phytoplankton uptake of the chemicals in question, and also changes in environmental concentrations, forms or species of the materials in question.

While seasonal effects are important, it appears that any changes are smaller than those found between polluted and clean areas (Cowan, 1981, Hummel et al, 1990). Once this variability is recognised it can be accommodated within the design of a field survey and experience suggests that responses to contamination may then be detected with a sensitivity appropriate to the demands of most monitoring programmes (Bayne, 1989).

Regardless of variation factors, mussels have an inherent variability in the population which cannot be attributed to another parameter. Baez and Bect (1989) suggest that this can be minimised by combining several individual organisms into one single sample to produce a better estimate of the mean concentration and variance of the pollutant. They found that samples containing 20 individuals showed the least variability. This backs up previous work by Bayne et al (1989) who considered that an optimum sampling size for mussels of between 15-25 individuals of different sizes was a requirement for monitoring biological effects.

While it seems unlikely that any one class of organism will satisfy all of the criteria required of an indicator organism, mussels possess many of the basic requirements, their biology is well understood and they have successfully been used in a number of studies (for example Goldberg 1978). In addition once a pattern of biomonitoring has been established, (which may take up to 3 consecutive years) there is no compelling reason to carry out annual assays, but monitoring at intervals of a small number of years may be adequate.

Polychaete Worms

Many organic contaminants may be found adsorbed to particulates suspended in the water column, and with time these sink to the benthos. Polychaetes make up 33-50% of the benthic macroinvertebrate fauna (Knox 1977) and since these species come into direct contact with the sediment, the potential effect of toxicants on polychaetes is of ecological importance.

Most lipophilic compounds which enter the marine environment rapidly become associated with sediment (Fowler et al 1978). It has been reported that there is a tendency for sediments to contain more residues that the overlying water in polluted areas. Knezovich et al (1987) indicate that these infaunal organisms are good choices for studies of chemical availability and sediment toxicity as they are likely to represent a "worst-case" for sediment exposures.

Despite the potential for high sediment concentrations of pollutants, relatively little attention has been focused on bioconcentration by benthic infauna (Shaw and Connell, 1987; Oliver, 1984; 1987, Connell et al, 1988). Where experimental studies have been conducted on the fate of pollutants, *Nereis virens* (king ragworm) has been one of the preferred marine species (Goerke and Ernst, 1977; 1986; Goerke, 1984b). It is a convenient size, has a lifespan of 3 years and can be maintained in laboratory conditions (Goerke 1979, 1984a). The closely related *Nereis diversicolor* (common ragworm) has also been used as it is more abundant, euryhaline but is a smaller animal and has a shorter life span of one to two years. The major pathway of uptake is uncertain. Fowler (1978) reported that *Nereis diversicolor* (common ragwom) obtained its pollutant burden from ingested sediment. However more recent studies by Shaw and Connell (1987), Oliver (1987) and McLeese et al (1980) indicate that the main route of uptake is from interstitial water. With *Nereis virens* accumulation, elimination and transformation of PCBs has already been studied in detail, on the assumption that results obtained in the laboratory can be related to natural conditions (Ernst et al 1977; Goerke and Ernst, 1977; Goerke, 1979; 1984a; 1984b). PCBs have been reported as persisting over a three week post exposure period in *Arenicola marina* (lugworm) and *Nereis diversicolor*.

There are factors which affect the accumulation and elimination of organic contaminants. Goerke and Weber (1990) found that while lipid content was not a major influence, differences in temperature and in population could significantly alter the elimination kinetics. Populations of *Nereis diversicolor* at a more polluted mesohaline site had a higher induction of degrading enzymes that the population from a polyhaline area. Rubinstein et al (1983) demonstrated that *Nereis virens* accumulated greater concentrations of PCB residues than did bivalves, for example, *Mercenaria mercenaria* (hard clam) or shrimps (for example *Palaemontes pugio),* and concluded that pathways in addition to direct uptake from water contributed to bioaccumulation. Despite their advantages, polychaetes have not been extensively used as biomonitors for organic contaminants in a field environment.

Common Dogwhelk

Nucella lapillus (common dogwhelk) has been used widely as a bioindicator of TBT contamination. While a number of other organisms have been used and found to bioaccumulate organotin compounds, the most sensitive responses discovered so far are those involving the reproductive impairment in gastropods from the sub-order Stenoglossa, such as *Nucella lapillus*. (Langston et al, 1990).

Gibbs et al (1987) offer reasons for the use of *Nucella lapillus* to monitor TBT as follows:

- It has a wide geographical distribution.
- Where it is found it is usually common, conspicuous and easily identified.
- It has a very limited potential for dispersal since its development is direct and the adults are slow moving.
- The eggs are laid in a conspicuous capsule which acts as a convenient marker of breeding activity.
- It is a hardy species, allowing for experimentation, such as transplantation to different areas.
- Most importantly it's sensitivity to TBT as manifested by imposex is seemingly unrivalled.

"Imposex" (Smith, 1971) describes the imposition of male characters, including penis and vas deferens onto the female. There are methods for quantifying long term changes in the intensity of imposex affecting a population, using Relative Penis Size (RPS) which is a relatively quick and easy method or Vas Deferens Stage (VDS) analysis which is a more time consuming method, but appears to be a more sensitive method of detecting low level TBT contamination (Bailey and Davies, 1988). As *Nucella lapillus* is a long lived species - six years or more, the level of imposex of adults can be considered to represent the response to long term exposure to TBT contamination. However it appears from studies by Gibbs et al (1987) that short term exposure levels can best be gauged by the degree of imposex in immature females that have yet to breed. Individual 12-18 months old seem to have the greatest potential for monitoring purposes. These authors were also able to show a significant relationship between tissue TBT concentration and the level of imposex observed in a field population of *Nucella lapillus*. There is no evidence that TBT induced imposex is fatal in the short term. In addition it is apparently an irreversible syndrome that is initiated by as little as 1 ng/l of TBT. Bryan et al (1986) reported that in south west England, populations of *Nucella lapillus* have been affected and the population is in decline. They suggest that the reduction in recruitment is caused by a lowered reproductive capacity rather than increased mortality rate. This is likely as the percentage of females in a population tends to fall as the degree of imposex increases and also it has been demonstrated that early stages in life cycle are most vulnerable. The decline in population is not masked by outside recruitment.

Other Bivalves

Langston (1978a,b) has performed extensive studies on the accumulation of Aroclors 1242, 1254 and 1260 by the marine bivalves *Macoma balthica* (Baltic tellin) and *Cerastoderma edule* (edible cockle). Both these species exhibited preferential accumulation of pentachlorobiphenyl isomers; isomers containing less chlorine atoms were accumulated in decreasing amounts. Langston (1978b) noted that the persistence of PCB isomers in the above species depends not only on the degree of chlorination but also on the position of the chlorine atoms in the molecule.

The uptake of PCB components by other bivalves, however, does not always follow this pattern. Thus although Nimmo et al (1975) found preferential accumulation of pentachlorobiphenyls in *Crassostrea virginica* (eastern oyster), Denton (1974) observed a greater persistence of lower chlorinated isomers in the clam *Mercenaria mercenaria* (hard clam), and Vreeland (1974) reported decreased accumulation of penta- to dichlorobiphenyls than of hexachlorobiphenyls in *Crassostrea virginica* (eastern oyster).

In Britain, TBT contamination has mainly been monitored through the use of *Nucella lapillus* (common dogwhelk), for example see Davies et al (1987), whilst in some areas extensive water analyses have been successful (Waldock et al, 1987). Scallops are also known to accumulate TBT and appear to have only limited ability to depurate following exposure (Davies et al, 1986). The accumulation of TBT in laboratory experiments has demonstrated high BCFs for *Ostrea gigas* (Pacific oyster) (Waldock and Thain, 1983) and for *Mytilus edulis* (common mussel) (Laughlin et al, 1986).

The 50% depuration rates for organic tin in bivalves are generally short, see table below.

Table 6.3 b   Depuration Rates for Organic Tin in Bivalves

Mytilus edulis	40 days	Zuolian and Jensen (1989)
Mytilus edulis	14 days	Laughlin *et al* (1986)
Ostrea edulis	10 days	Waldock *et al* (1983)

Several studies (Alzieu et al, 1976; Waldock and Miller, 1983) have demonstrated the accumulation of TBT in natural oyster populations. Zuolian and Jensen (1989) have demonstrated that, in *Mytilus edulis* (common mussel), organotin uptake is rapid and that accumulation rate increases exponentially with decreasing concentrations of organotin in seawater. This relationship in bivalves has also been observed by Salazar et al (1987), Waldock et al (1983) and Unsal (1984).

In a study of tin and organotin in Poole Harbour, Dorset, Langston et al (1987) were able to demonstrate that various benthic invertebrates were useful as indicators of organotin contamination.

Slow metabolism of TBT has been reported for *Crassostrea virginica* (eastern oyster), (Lee,1986) and this may account for the exceptionally high levels of organotin, particularly TBT, observed by Langston et al (1987) in the clam, *Mya*. The high BCFs observed in the sediment-dwelling bivalves *Scrobicularia plana* and *Mya arenaria* suggest particulate bound organotins may be a source of contamination in addition to water.

6.3.3 Saltwater Fish

A variety of fish species has been used as indicator species for chlorinated hydrocarbons in the Thames Estuary (Rickard and Dulley 1983), the Medway Estuary (Wharfe and van den Broek, 1978; van den Broek, 1979) and the North Sea (Buther, 1990; Knickmeyer and Steinhart, 1989).

The MAFF Directorate of Fisheries Research use utilise six fish species as "core" indicator species for trace metals, organochlorine insecticides and PCBs, These are given in Table 6.3c.

Table 6.3 c

MAFF Directorate of Fisheries Research Fish Indicator Species
Roundfish
Cod (*Gadus morhua*)
Whiting (*Merlangius merlangus*)
Flatfish
Plaice (*Pleuronectes platessa*)
Sole (*Solea solea*)
Dab (*Limanda limanda*)
Flounder (*Platicthys flesus*)

In addition to these *Osmerus eperlanus* (smelt) has been reported as being useful since it is nektonic and thus occupies a different niche from most other fish used, which tend to be mainly benthic (Rickard and Dulley, 1983).
The levels of most chlorinated hydrocarbons in fish are generally greater in the liver than in the muscle tissue (Rickard and Dulley, 1983). However since muscle tissue in most cases accounts for at least 90% of the total body weight, this has also been studied for pollutant levels. Wharfe and van den Broek (1978) found significant correlations between hexane extractable fat and dieldrin levels in muscle tissue of a variety of fish species. Lipid content may be an important factor determining the actual levels accumulated by fish but the effect of this does vary. For example since gamma-HCH is less lipophilic than most organic

compounds (Gunther et al, 1968) its tissue levels in fish shows more age dependence than lipid content dependence (Rickard and Dulley, 1983). The use of fish in monitoring certain organochlorines can be advantageous since the clearance of accumulated PCBs from the body is slow (Bengtsson, 1980; Defoe et al, 1978; Mayer et al, 1977). The high levels of PCBs *Solea solea* (sole) for example enabled the pinpointing of a contamination source in the vicinity of Tilbury (Rickard and Dulley, 1983).

Individual species of fish vary greatly and this should be considered in any attempts at comparisons between species. Buther (1990) found that the mean *Limanda limanda* (dab) liver content of contaminants (22%) in December 1984 was twice as high as that of *Platicthys flesus* (flounder) liver (10%) in September 1988 in the Wadden Sea. *Platicthys flesus* (flounder) and *Pleuronectes platessa* (plaice) which are closely related species with similar diets also displayed differences (van den Broek, 1979). On average the chlorinated hydrocarbon concentrations were 65% lower in the latter species. It has also been noted with studies of different species of fish that the distribution of PCB congeners among tissues is species-dependent (Hansen et al, 1971; Guiney and Peterson, 1980).

Another factor that needs to be considered is that fish possess mixed function oxygenase (MFO) enzyme systems which are capable of degrading some organic contaminants. An extra complication may arise therefore that if, for example, the concentration of PCB in the liver of *Solea solea* (sole) reached a sufficiently high level it would act to induce hepatic MFO activity (Stein et al, 1984).

Mouvet et al (1985) have argued that fish may not be considered very good bioindicators since they do not possess all of the desired indicator characteristics, namely, they are:

- not sedentary;
- often difficult to sample;
- do not always tolerate high levels of toxicants;
- their accumulation capacity may vary considerably according to the season,species, age, sex, and organ sampled (Olsson and Jensen, 1978).

7 GENERAL DISCUSSION

7.1 Introduction

This review has attempted to bring together as much of the relevant research data as is currently available on the topic of bioaccumulation as an approach for detecting and monitoring Red List trace organic compounds. The review has firstly looked at the characteristics of the specific groups of compounds with reference to their bioaccumulation potential. It has then looked at the mechanisms involved, the advantages of the approach, and factors affecting the reliability of organisms. The preceding section then looked at current usage of various different groups of indicator organisms in both fresh and estuarine / coastal environments.

This section aims to set out a series of general comments which refer back to the original objectives of the review, namely to consider biaccumulation in terms of its advantages, its applicability, the levels of confidence in it and how best to interpret the data.

7.2 Advantages

These have been dealt with in some detail in Section 4 but it is considered appropriate here to summarise the broader implications.

- There is no doubt that bioaccumulation offers many advantages in the detection of contamination by certain groups of Red List trace organics. In particular this can be seen with the more lipophilic non-polar compounds.

- Measuring tissue concentrations in sedentary organisms provides distinct advantages in detecting pollutants in both freshwaters and estuaries or coastal waters. Thus in rivers it can be an effective method for picking up intermittent contamination and in estuaries a way of locating potential hot-spot areas.

- The fact that certain organisms are able to take up and retain various trace organic compounds with minimal, if any, metabolic transformation of the toxicant means that:

  - the body burden reflects levels of environmental contamination, and

  - pollutants can be identified following an episodic event after they have disappeared from the water column.

- Bioaccumulation offers a further dimension to detecting and monitoring these compounds since it gives an indication of their bioavailability and hence contributes to an understanding of their impact on the environment.

- Although the sophistication of instruments used to measure trace organics in water is continuing to evolve and thus limits of detection for specific compounds are tending to come down, bioconcentration still offers an advantage in making detection easier through this "natural" concentration step.

- There are attractions in using simpler organisms for wider application as potential monitors of the Red List compounds. In particular these would include aquatic mosses and possibly some macroalgae in freshwaters and macroalgae in estuarine systems. These organisms have, however, been largely ignored for this application until recently so method development and interpretation of the data needs further advances and practice before it can be routinely adopted.

7.3 Applicability

- Bioaccumulation as an approach for the detection and monitoring of compounds does not appear to be applicable to two of the main groups of Red List trace organics substances, namely the organophosphorus compounds and the organic solvents. Organophosphorus compounds appear to degrade too quickly through hydrolysis and oxidation, producing water soluble products which are simply not taken up by organisms to an extent which makes it an attractive method for detection and monitoring. Organic solvents appear to be too volatile and unstable in the aquatic environment for appreciable if any bioaccumulation to take place.

- Bioaccumulation as an approach is applicable for the organochlorine compounds (including PCBs), the organotin compounds and some of the more environmentally important herbicides.

- In terms of monitoring trends in the concentrations of trace organic pollutants great care needs to be taken with respect to the use of bioaccumulation as an approach. A number of important factors influence how the approach may be applied including:

    - choice of indicator organism and

    - which fraction of the pollutant in the environment is the focus of the monitoring programme (eg. the fraction in the water, associated with particulates, or associated with sediments).

- It has been shown that the use of higher organisms in particular is complicated by a number of factors - age, season, diet, behaviour etc which all need to be considered and evaluated to permit any accurate estimation of the pollution conditions and in turn the subsequent trends in these conditions.

- Simpler organisms which lack many of these complicating features would appear to lend themselves better to monitoring trends in organic contaminant concentrations. The only drawback to this though is that the majority of these organisms have received very little attention so there is the need for more developmental work in order that they can be more widely applied.

- There are attractions in the potential use of plants in both fresh and estuarine waters (see 4.1.3 for a discussion of the advantages) but their application to detection and subsequent monitoring of trends has only been comparatively recently recognised. There is a lack of a large database currently on which to base their use as monitors.

7.4 Levels of Confidence

- Concern has been expressed in the levels of confidence that can be attributed to bioaccumulation survey results especially involving higher organisms where the factors reviewed in Section 5 (including sex, age, diet, seasonality etc) can influence tissue concentrations. These factors have received a lot of attention particularly with respect to organisms such as the mussel, and so long as controls on the sampling of organisms and the standardization of methods are carried out then results with acceptable standard deviations and limits of confidence can be achieved.

- Recent trends in the advancement of instrumentation for the analysis of trace organic contaminants coupled with more sophisticated techniques for extraction, clean-up and concentration mean that increasing confidence can be placed in tissue analysis.

7.5 Interpretation

In its simplest application the measurement of tissue concentrations of trace organic pollutants indicates:

- their presence in the environment;

- their potential bioavailability;

- their potential for bioconcentration;

- their potential for biomagnification within a food chain.

- In situations where pollutants are released intermittently into the environment and their presence in the water column is of relatively short duration but their ecological impact can be substantial , then bioaccumulation by sedentary organisms can again lead to both the detection of the compound and pin-point the source.

- The application of bioaccumulation of trace organic compounds does not appear to be sufficiently advanced as yet to enable the accurate quantification or prediction of external concentrations of contaminants in the water column. This may in part be due to the focus of attention having been placed on higher organisms where bioaccumulation is influenced and clearly complicated by a variety of different factors such as size, age, season, diet, metabolic functions, behavioural responses etc. All of these factors complicate interpretation and necessitate a great deal of control in the selection of organisms and the standardization of methodologies.

- From the literature it does appear that bioaccumulation can be applied to monitoring trends in the levels of organic contaminants. The only appropriate organisms, however, are those which have received sufficient research into their bioaccumulation capacity and where detailed studies on background concentrations as well as elevated concentrations under polluted conditions have been carried out. There are, however, few organisms which have received such detailed and rigorous study particularly with reference to the wide range of compounds in the Red List of trace organics. In coastal and estuarine systems probably *Mytilus edulis* (common mussel) is one of the few organisms which has received sufficient study (Mussel Watch Programmes) and this is only with reference to organochlorine pesticides and PCBs.

- Current research on the interpretation of bioaccumulation data is looking at combining bioaccumulation with physiological response measurements for higher organisms. Thus for *Mytilus edulis* (common mussel) recent research has looked at establishing laboratory derived relationships between the concentration of trace organic toxicants in tissues and sublethal physiological responses, in order to assess and monitor pollution by certain compounds (Widdows and Donkin, 1989).

8 SUMMARY CONCLUSIONS

Bioaccumulation as an approach to detecting monitoring pollution by Red List trace organic compounds will not replace chemical monitoring but should be adopted in some cases as a complimentary and in others as a supplementary approach.

- There is unlikely to ever be a universal indicator organism for either freshwater or estuarine/coastal environments because of the variable source of polluting compounds (water soluble, particulate-bound, sediment-bound) and the variability of the particular environments (flow regimes, substrate, salinity gradients etc).

- Bioaccumulation is applicable to organochlorine and organotin compounds and probably some chlorinated herbicides. It does not appear to be applicable to organophosphorus and organic solvent compounds.

- The applciation of bioaccumulation monitoring is not sufficiently advanced to enable the accurate quantification or prediction of external concentrations of trace organic contaminants in the water column.

- It will be necessary to standardise methodologies for a variety of different indicator organisms in order to apply the bioaccumulation approach to detecting and monitoring trace organic compounds in the range of water bodies that will be of interest to the NRA.

Annex 1

Current Usage of Indicator Organisms for Bioaccumulation Monitoring by the NRA

Freshwater Species

Species	Utilized by	Advantages	Disadvantages
Eel *(Anguilla anguilla)*	NRA (5 regions) MAFF DoE	* High lipid content. *Widespread occurrence. * Easy to catch. * Relevant to public health issues.	* Highly mobile - amphibious. * Migratory species. *Methodological problems associated with the use of complex organisms such as fish.
Gudgeon *(Gobio gobio)*	NRA (Yorkshire region)	* Common in polluted rivers. * Relatively sessile.	* Methodological problems associated with the use of complex organisms.
Trout *(Salmo trutta)*	NRA (Yorkshire region)	* Widespread in upper reaches of systems. * Relevant to public health issues.	* Not found through-out river systems. * Methodological problems as above.

Species	Utilized by	Advantages	Disadvantages
Moss *(Rynchostegium riparioides)*	NRA (Yorkshire region) NECL (1989)	* Sedentary. * Morphology minimises problems due to different-iated organs. * Pollution tolerance. * Large surface area. * No excretion mechanisms.	* Presumably not responsive to particulate phase of organics. * Distribution problems.

Estuarine Species

Species	Utilized by	Advantages	Disadvantages
Common Mussel *(Mytilus edulis)*	NRA (8 regions) MAFF JMG (Primary choice species) RPBs (2)	* Presumably responsive to particulate and dissolved phases of organics. * Sedentary. * Required for shellfish directive work. * Widely employed.	* Distribution problems in many estuaries; restricted to more marine sites and rocky shores. * Methodological problems associated with higher organisms (eg. gut clearance etc).
Oyster *(Ostrea edulis)*	NRA (3 regions)	* Presumably responsive to particulate and dissolved phases of organics. * Required for shellfish directive.	* Distribution problems, commoner in southern coasts. * Methodological problems.
Zebra Mussel *(Dreissana spp)*	NRA (Thames)	* Presumably responsive to particulate and dissolved phases of organics. * Found in lower salinity range.	* Restricted distribution. * Methodological problems.

Species	Utilized by	Advantages	Disadvantages
Cockle *(Cardium edule)*	NRA (Wessex)	* Presumably responsive to particulate and dissolved phases of organics. * Relevant to public health issues in some areas.	* Methodological problems.
Periwinkle *(Littorina littorea)*	NRA (2 regions)	* Usually obtainable in large numbers.	* Methodological problems. * Restricted to rocky shores.
Limpet *(Patella spp)*	NRA (3 regions)	* Very restricted home range ie. almost sedentary.	* Methodological problems. * Restricted to rocky shores.
Ragworm *(Nereis diversicolor)*	NRA (Yorkshire)	* Found in wide range of salinities. * Presumably responsive to sedimentary phase of organics.	* Can be time consuming to get large enough sample. * Restricted to more marine sites on estuaries and confined to silty/muddy areas.
Shrimp *(Crangon crangon (vulgaris))*	NRA (Severn - Trent)	* Presumably responsive to dissolved and particulate phases of organics.	* Restricted distribution. * Methodological problems.

Species	Utilized by	Advantages	Disadvantages
Dab *(Limanda limenda)*	NRA (2 regions) JMG (primary choice species) NSTF MAFF	* Liver with high lipid content. * Interface with NSTF monitoring work.	* Methodological problems associated with complex organisms such as fish. * Migratory. * Mobile.
Flounder *(Platichthys flesus)*	NRA (4 regions) JMG (2° species) MAFF	* Liver with high lipid content.	* Methodological problems as above. * Migratory. * Mobile.
Sole *(Solea solea)*	MAFF	* As above.	* As above.
Plaice *(Pleuronectes platessa)*	MAFF	* As above.	* As above.
Cod *(Gadus morhua)*	NRA (Thames) MAFF	* As above.	* As above.
Wracks *(Fucus Spp.)*	NRA (7 regions)	* Sedentary.	* Not widespread in all estuaries. * Presumably only responsive to dissolved phase of trace organics. * Methodological conflicts as to which part of the thallus to sample.

Species	Utilized by	Advantages	Disadvantages
Enteromorpha spp.	NRA (3 regions)	* Sedentary. * Found in lower salinity zone of estuaries. * Ubiquitous.	* Presumably only responsive to dissolved phase of trace organics.
Sea lettuce *(Ulva lactuca)*	NRA (Severn Trent)	* Sedentary.	* Distribution problems. * Methodological problems.
Red Algae *(Chrondus crispus, Ceramium spp.)*	NRA (Severn Trent)	* Sedentary.	* As above.

Eel Methods

Authority	Sampling	Sample Preparation	Analysis	Red List Organics Analysed For
Wessex NRA	10 eels sampled per site. 5g muscle fillet and whole liver sampled.	Homogenised and deep frozen to wait analysis	GC-ECD	DDT,HCBD, lindane, HCB, drins, endosulfan, PCB.
Yorkshire NRA	No set programme of sampling. Muscle tissue sampled and deep frozen.	Freeze dried and pulverised. Soxhlet extraction with hexane for 2 hours. Silica column clean - up.	GC-ECD	DDT,lindane, drins, PCB.
Northumbria NRA	10 eels of 12 inches length sampled per site. Individual fish analysed. Muscle tissue. Deep frozen.	Follow Determination of Organochlorine Insecticides and PCBs (1984) method.	GC-MS	DDT,lindane, drins, PCB.
Severn - Trent NRA	Sampling once a year. Muscle tissue.	Homogenised, dried with sodium sulphate. Extracted with solvent. Column clean - up.	GC-ECD	DDT,lindane, dieldrin, aldrin, endosulfan.

Authority	Sampling	Sample Preparation	Analysis	Red List Organics Analysed For
Welsh NRA	Liver and muscle sampled (sometimes gill also).	Homogenised. Soxhlet extraction with hexane (1-2 hours). Aluminium oxide clean - up column.	GC-MS	DDT (and metabolites), lindane,endosulfan,drins PCB.

Estuarine Fauna Methodologies: Organism Sampling

Organism	No of individuals per sample	Size Range (mm)	Season of collection	Demonstrated Bioaccumulation For	Utilised by
Mytilus edulis	50	45 - 55	July - Aug Jan - Feb	DDT, Dieldrin, Lindane, Aldrin, Endosulfan PCBs. -	Welsh Water Authority (No TW82/13).
	50	25-45	Mid Jan - Mid Mar		NRA Baseline Estuary and coastal waters Monitoring Programme.
Nereis diversicolor	100	N/A	Aug/Sept	-	NRA Baseline.
Littorina littorea	50	>10 diameter	July - Aug Jan - Feb	DDT, Dieldrin, Lindane, PCBs. -	Welsh Water Authority (No TW82/13).
	30	c.20	Aug/Sept Nov		NRA Baseline.
Patella spp	50	>30 diameter	July - Aug Jan - Feb	DDT, Dieldrin, HCH, aldrin, PCBs.	Welsh Water Authority (No TW82/13).
	50	March - May	c. 40 dia		NRA Baseline.

Species	Storage Prior to Cleansing/ Preparation	Cleansing	Depuration	Storage Prior to Dissection	Tissue Selection	Storage Prior to Analyses	Authority
Mytilus edulis	No	Washed in tap water.	24 h in aerated seawater.	Frozen 18°C.	Whole soft tissues.	No	Welsh Water Authority (No TW82/13).
	No	Scrape off growth on shells and scrub clean.	48 h in clean water.	Deep frozen.	Remove shells.	Can be frozen but best analysed immediately.	NRA Baseline Estuary and coastal waters Monitoring Programme.
Nereis diversicolor	Kept cool in sediment (up to 24 hrs).	Gentle washing (in fine sieve).	6 days in acid - washed sand, 1 day in clean water only.	Deep frozen.	Whole animals.	Equivalent to storage prior to dissection.	NRA Baseline.
Littorina littorea	-	Washed with distilled water.	-	-	Soft tissues.	Frozen -18°C.	Welsh Water Authority (No TW82/13).
	No	Thorough washing in clean water.	-	Deep frozen.	Remove shells after boiling for 1 min or steaming.	Can be frozen but best analysed immediately.	NRA Baseline.
Patella spp	-	Washed with distilled water.	-	-	Soft tissues.	Frozen-18°C.	Welsh Water Authority (No TW82/13).
	No	Thorough washing in clean water.	-	Deep frozen.	Remove shells.	Can be frozen but best analysed immediately.	NRA Baseline.

Species	Storage Prior to Cleansing/ Preparation	Cleansing	Depuration	Storage Prior to Dissection	Tissue Selection	Storage Prior to Analyses	Authority
Limanda limanda *Platichthys flesus*	Refrigeration (up to 24 hrs).	Thorough washing and gentle scrubbing, remove mucilage and attached matter.	-	Deep frozen.	Remove liver.	Can be frozen but best analysed immediately.	NRA Baseline.

Sample	Extraction Method	Clean - up Solvent	Column	Eluant	Red List Trace Organics Analysed	Analysis	Authority
Mytilus edulis, Littorina littorea, Patella spp	Soxhlet	n - hexane	Alumina/ silica gel	n - hexane	Lindane, drins, DDT, PCBs Endosulfan.	GC-ECD	Welsh Water Authority (no TW82/13).
Mytilus edulis, Gadus morhua, Merlangius merlangius, Pleuronectes platessa, Solea solea, Limanda limanda, Platichthys flesus.	Soxhlet	n - hexane	Alumina/ silica	n - hexane	HCB lindane, dieldrin DDT + metabolites, PCBs.	GC-ECD	MAFF (1990)
Fish Tissues	Soxhlet (2 -3 hours)	Hexane	Alumina/ silver nitrate	Hexane	-	GC-ECD	Determination of Organochlorine Insecticides and PCBs (1984).

Fucus and Enteromorpha Methods

Authority	Species	Sampling	Sample Preparation	Analysis	Red List Organics Analysed For
Welsh Water Authority (Tidal Waters Report No TW82/13)	*Fucus vesiculosus*	Jan - Feb 1 kg sample. Below mid-tide level.	Washed 3 times in tap water to remove mucus and other material, rinsed in distilled water. Whole fronds frozen at -18°C. Stems and/or tips dissected from 25 fronds and ground. Soxhlet extraction with n - hexane. Alumina/silica clean-up. Eluted with n - hexane.	GC-ECD	Lindane, drins, DDT (and metabolites).
Severn Trent NRA	*Fucus vesiculosus* *F.serratus* *Enteromorpha spp*	4 times/year minimum of 6 plants from same mid-tide position.	Washed repeatedly. Samples frozen. Homogenised or ground up, dried with sodium sulphate. Solvent extraction column clean-up.	GC-ECD	DDT, lindane, dieldrin, aldrin, endosulfan.
Southern NRA	*Fucus vesiculosus* or *F.serratus*	Annual sampling in Feb/March.	Soxhlet extraction, follow MAFF published procedures.	GC-ECD	DDT (and metabolites), lindane, drins, PCBs.

Authority	Species	Sampling	Sample Preparation	Analysis	Red List Organics Analysed For
Yorkshire NRA	*Fucus vesiculosus* *Enteromorpha spp*	Biannual. *Enteromorpha* taken from available tide level. *Fucus* collected at mid-tide level.	*Enteromorpha* prepared according to method of Say & Burrows (1985). Whole fronds used. Wet weight C 15-20g. *Fucus*. New tissue (distal 5 cm) and fruiting bodies discarded. Flat thallus between bladders is removed for analysis. C 15-20g. All samples frozen, dried, pulverised. Soxhlet extraction with hexane for 2 hours. Silica column clean-up.	GC-ECD	Drins,lindane, DDT,PCBs.
Anglian NRA	*Fucus spp*	As above.	As above.	GC-ECD	Not decided.
Wessex NRA	*Fucus serratus* *F. vesiculosus*	Annual.	Washed in distilled water. Frozen,thawed, tips of fronds removed and homogenised. Refrozen to await analysis.	GC	DDT, HCBD, lindane, drins, endosulfan, PCBs.
Thames NRA	*Enteromorpha spp* *Fucus spp*	As per guidance in NRA Bioaccumulation protocol. Sampling annually.	Follow guidance in NRA Bioaccumulatioin Protocol and MAFF Analytical Publications.	GC-ECD	DDT, HCBD, PCP, lindane, HCB, Drins, atrazine, simazine.
Welsh NRA	*Fucus vesiculosus*	1 kg stems and/or thallus, tips dissected out .	Washed 3 times in tap water, rinsed in distilled water and frozen at -18 °C. Bulked samples ground/homogenised. Extraction using hexane.	GC-MS	DDT (and metabolites), lindane, drins, PCBs, endosulfan.

Authority	Species	Sampling	Sample Preparation	Analysis	Red List Organics Analysed For
NRA Baseline Estuary and Coastal Waters, Monitoring Programme	*Fucus spp* preferably *F.serratus*	25-30 plants from mid-shore in size range 250 - 300 mm. February.	Refrigeration up to 10 days. Scrubbing and washing. Select old thallus only.	GC	Not stated.

Annex 2

References

Adams WJ, Kimerle RA and Mosher RG (1983) **Aquatic Safety Assessments of Chemicals Sorbed to Sediments**, presented at ASTM Seventh Annual Aquatic Toxicology Symposium, April 17-19, Milwaukee, Wisconsin

Addison RF, Zinck ME and Leahy JR (1976) **Metabolism of Single and Combined Doses of ^{14}C aldrin and ^{3}H-p,p'-DDT by Atlantic Salmon (*Salmo salar*) fry**, J Fish Res Bd Canada 33, 2073-76

Addison RF and Zinck ME (1977) **Rate of Conversation of ^{14} C-p,p'-DDT to p,p'-DDE by brook trout (*Salvelinus fontinalis*): Absence of Effect of Pretreatment of Fish with Compounds Related to p,p'-DDT**, J Fish Res Bd, Canada 34, 119-22

Alzieu Cl, Sanjuan J, Deltreil JP and Bovel M (1986) **Tin Contamination in Archachon Bay: Effects on Oyster Shell Abnormalities**, Mar Pollut Bull, 17, 494-498

Anderson RB and Everhart WH (1966) **Concentrations of DDT in Landlocked Salmon (*Salmo salar*) at Sebago Bay, Maine**, Am Fish Soc, 95, 160-164

Anderson RB and Fenderson OC (1970) **An Analysis of Variation of Insecticide Residues in Landlocked Atlantic Salmon (*Salmo salar*)**, J Fish Res Bd Canada 27, 1-11

Anderson J R and Orbaek K (1984) **Organochlorine Contaminants in Human Milk in Denmark, 1982**, Ambio Vol 13 No 4

Anderson R L and Defoe D L (1980) **Toxicity and Bioaccumulation of Endrin and Methoxychlor in Aquatic Invertebrates and Fish**, Environ Pollut Ser A 22, 111 - 121

Aubert J, Flatau G, Puel D, Breittmayer V and Clement R (1977) **Etude de la Contamination des Élevages Marins**, Rev Int Oceanogr Med 45-46, 77-97

Baez F B P and Bect G M S (1989) **DDT in *Mytilis edulis* : Statistical Considerations and Inherent Variability**, Mar Pollut Bull 20, 496 - 499

Bailey S K and Davies I M (1987) **Tributyltin Contamination in the Firth of Forth**, Proc Roy Soc Edin, 93B, 561 - 562

Bailey S K and Davies I M (1988) **Tributyltin Contamination around an Oil Terminal in Sullum Voe (Shetland)**, Environ Pollut 55, 161 - 172

Barber M C, Lassiter R R and Suarez L A (1988) **Modelling Bioconcentration of Non-polar Organic Pollutants by Fish**, Environ Toxicol Chem 7 (7), 545 - 588

Barber M C, Suarez L A and Lassiter R R (1991) **Modelling Bioaccumulation of Organic Pollutants in Fish with an Application to PCBs in Lake Ontario Salmonids**, Can J Fish Aquat Sci 48 (2), 318 - 337

Barcelo D (1991) **Occurrence, Handling and Chromatographic Determination of Pesticides in the Aquatic Environment - A Review** Analyst 116 (7), 681 - 689

Barnett B E and Ashcroft C R (1985) **Heavy metals in *Fucus Vesiculosus* in the Humber Estuary,** Environ Pollut Ser B 9, 193 - 213

Bauer I, Weigelt S and Ernst W (1989) **Biotransformation of Hexachlorobenzene in the Blue Mussel (*Mytilus edulis*),** Chemosphere 19 (10 - 1), 1701 - 1707

Bayne BL (1976) (Ed) **Marine Mussels: Their Ecology and Physiology.** Cambridge University Press, Cambridge, pp 506

Bayne B L (1989) **Measuring the Biological Effects of Pollution: The Mussel Watch Approach** Wat Sci Technol 21, 1089 - 1100

Bayne BL, Brown DA, Harrison F and Yevich PD (1980) **Mussel Health,** see National Academy of Sciences, 1980

Bedford JW, Roelofs EW and Zabik MJ (1968) **The Freshwater Mussel as a Biological Monitor of Pesticide Concentrations in a Lotic Environment,** Limnol Oceanogr, 13, 118-26

Bedford J W and Zabik M J (1973) **Bioactive Compounds in the Aquatic Environment: Uptake and Loss of DDT and Dieldrin by Freshwater Mussels,** Arch Environ Contam Toxicol 1, 97 - 111

Bellar TA, Lichtenberg JJ and Lonneman SC (1980) in RA Baker (Ed) **Contaminants and Sediments,** Vol 2, Analysis, Chemistry, Biology, Ann Arbor Science Publishers Inc / The Butterworth Group, Ann Arbor, MI, 57-70

Bengtsson B-E, (1980) **Long Term Effects of PCBs (Clophen A50) on Growth, Reproduction and Swimming Performance in the Minnow,** *Phoxinus phoxinus,* Wat Res 14, 681-687

Bertrand P, Fournier S and Vigneaulty Y (1986) **Concentrations en Biphenyles Polychlorés et en Métaux dans les Sédiments de la Baie des Anglais (Québec),** Rapp Can Sci Halieut Aquat 568, pp 5

Biggar JW, Doneen LD and Riggs RL (1966) **Soil Interaction with Organically Polluted Water,** Summary Report, Davies, California, Department of Water Science and Engineering, University of California

Bjerk J E and Brevik E M (1980) **Organochlorine Compounds in Aquatic Environments,** Arch Environ Contam Toxicol 9, 743 - 50

Boehm PD and Quinn JG (1976) **The Effect of Dissolved Organic Matter in Seawater on the Uptake of Mixed Individual Hydrocarbons and Number 2 Fuel Oil by a Marine Filter-Feeding Bivalve *(Mercenaria mercenaria),*** Estuar Cstle Mar Sci, 4, 93-105

Boëtius I and Boëtius J (1985) **Lipid and Protein Content in *Anguilla anguilla* During Growth and Starvation,** Dana 4, 1-18

Böhm HH (1976) **Sorption und Wirkung von Chlortriazin-Phenoxyfeltsäuren - und Bipyridiliumherbiziden in Statischen und Kontinuierlichen Kulturen Planktischer Blau-, Kisel - und Grünalgen -** Dissertation Universität Hohenheim 128 S

Borlakoglu J and Dils R (1990) **Polychlorinated Biphenyls (PCBs) and Marine Food Chains**, Biologist 37 (5)

Boryslawskyj M, Garrood A C and Pearson J T (1987) **Rates of Accumulation of Dieldrin by a Freshwater Filter Feeder:** *Sphaerium corneum*, Environ Pollut 43, 3 - 13

Boryslawskyj M, Garrood T, Stanger M , Pearson T and Woodhead D (1988) **Role of Lipid / Water Partitioning and Membrane Composition in the Uptake of Organochlorine Pesticides into a Freshwater Mussel**, Mar Environ Res 24, 57 - 61

Bowmer KH (1987) **Herbicides in Surface Water**, In: Roberts TR and Hutson D (Eds) Herbicides. Progress in Pesticide Biochemistry and Toxicology Vol 6, John Wiley and Sons, New York

Boyden CR (1977) **Effect of Size upon Metal Content of Shellfish**, J Mar Biol Assoc UK 57, 675-714

Brodtmann NV (1970) **Studies on the Assimilation of 1,1,1-trichloro-2,2-bis (p-chlorophenyl)ethane (DDT) by** ***Crassostrea virginica*** Gmelin, Bull Environ Contam Toxicol 5, 455-62

Brown L (1980) **The Use of** *Hydrobia jenkinsi* **to Detect Intermittent Toxic Discharges to a River**, Wat Res Vol 14, 941 - 947

Brusle J (1990b) **L'anguille Européenne (Anguilla anguilla), une Espèce Jugée Commune Jusqu'a la Dernière Dècennie Mérite-t-elle d'acquérir Aujourd'hui le statut d'espèce menacée?** Bull Soc Zool Fr, 114, 61-73

Brusle J (1991) **The Eel (Anguilla spp) and Organic Chemical Pollutants**, Sci Tot Environ 102, 1 - 19

Bryan G W (1969) **The Absorption of Zinc and Other Metals by the Brown Seaweed** ***Laminaria digitata***, J Mar Biol Ass Uk 49, 225, 243

Bryan G W (1979) **Bioaccumulation of Marine Pollutants**, Philos Trans R Soc Lond B Biol Sci 286, 483 - 505

Bryan G W, Gibbs P E, Hummerstone L G and Burt G R (1986) **The Decline of the Gastropod** *Nucella lapillus* **around South-west England: Evidence for the Effect of Tributyltin from Antifouling Paints**, J Mar Biol Ass UK 66, 611 - 640

Buhler DR, Rasmusson ME and Shanks WE (1969) **Chronic Oral DDT Toxicity in Juvenile Coho and Chinook Salmon**, Toxicol Appl Pharmacol 14, 535-55

Buhler DR and Shanks WE (1970) **Influence of Body Weight on Chronic Oral DDT Toxicity in Coho Salmon,** J Fish Res Bd, Canada 27, 347-58

Bulter PA (1969a) **The Significance of DDT Residues in Estuarine Fauna, In: Chemical Fallout** (MW Miller and Berg GG (Eds)), pp 205-20, Springfield, Illinois, Charles C Thomas

Bulter PA (1969b) **Monitoring Pesticide Pollution,** BioScience 19, 889-91

Bulter PA (1971) **Influence of Pesticides on Marine Ecosystems,** Proc R Soc Lond B 177, 321-9

Burnett R (1971) **DDT Residues: Distribution of Concentrations in *Emerita analoga* (Simpson) along Coastal California,** Science 174, 606-8

Buser H-R (1990) **Atrazine and Other S-triazine Herbicides in Lakes and in Rain in Switzerland,** Environ Sci Technol 24 (7), 1049 - 1058

Buther H (1990) **Spatial and Temporal Trends in Organochlorine Contamination of Dab (*Limanda limanda*) and Flounder (*Platichthys flesus*) in the North Sea,** Archiv fur Fischereiwissenschaft 40 (1-2), 133 - 152

Butler G L (1977) **Algae and Pesticides,** Residue Rev 19, 19 - 62

Butler P A (1973) **Organochlorine Residues in Estuarine Molluscs 1965 - 1972. National Pesticides Monitoring Program,** Pestic Monit J 6, 238 - 362

Butler PA, Andren L, Bonde GJ, Jernelou A and Reisch DJ (1971) **Monitoring Organisms, In: FAO Technical Conference on Marine Pollution and its Effects on Living Resources and Fishing, Rome 1970, Supplement 1: Methods of Detection, Measurement and Monitoring of Pollutants in the Marine Environment,** Ed M Ruivo, 101-12, London, Fishing News (Books) Ltd

Calambokidis J, Mowrer J, Beug M W and Herman S G (1979) **Selective Retention of Polychlorinated Biphenyl Components in the Mussel, *Mytilus edulis*,** Arch Environ Contam Toxicol 8, 299 - 308

Canton J W, Greve P A, Sloof W and van Esch G J (1975) **Toxicity, Accumulation and Elimination Studies of alpha-hexachlorocyclohexane (alpha-HCH) with Freshwater Organisms of Different Trophic Levels,** Water Res 9, 1163 - 1169

Canyurt MA (1983) **Action toxique du Lindance et du Parathion Methyl sur Troi Espèces de Poissons d'eau douce,** Bull Cent Etud Rech Sci Biarritz 14 (3), 257-162

Carter C W and Suffet I H (1982) **Binding of DDT to Dissolved Humic Materials,** Environ Sci Technol 16, 735 - 740

Chadwick GF and Brocksen RW (1969) **Accumulation of Dieldrin by Fish and Selected Fish-Food Organisms,** J Wildl Man 33, 693-700

Chiou CT, Peters LJ and Freed VH (1979) **Science**, 206, 831

Chiou CT, Porter PE and Schmedding DW (1983) Environ Sci Technol, 17, 227

Chisholm DA, MacPhee W and MacEachem CR (1955) **Effects of Repeated Applications of Pesticides to Soil,** Can J Agric Sci, 35, 433

Cleary J J and Stebbing A R D (1985) **Organotin and Total Tin in Coastal Waters of South-west England,** Mar Pollut Bull, 16, 350 - 355

Cole H, Barry D, Frear DEH and Bradford A (1967) **DDT Levels in Fish, Streams, Stream Sediments and Soil Before and After DDT Aerial Spray Application for Fall Cankerworm in Northern Pennsylvania,** Bull Environ Contam and Toxicol 2, 127-46

Connell D W (1988) **Bioaccumulation Behaviour of Persistent Organic Chemicals with Aquatic Organisms,** Reviews of Environ Contam Toxicol, Vol 101, 117 - 147

Connell D W, Bowman M and Hawker D W (1988) **Bioconcentration of Chlorinated Hydrocarbons from Sediment by Oligochaetes,** Ecotoxicol and Environ Safety 16, 293 - 302

Connor MS (1983) **Fish / Sediment Concentration Ratios for Organic Compounds,** Environ Sci Technol, 18, 31-5

Connor MS (1984) Environ Sci Technol, 18, 31

Conte FS and Parker JC (1975) **Effect of Aerially-Applied Malathion on Juvenile Brown and White Shrimp** ***Penaeus aztecus*** **and** ***P setiferus*** Trans Amer Fish Soc 104, 793-9

Cook GH, Moore JC and Coppage DL (1976) **The Relationship of Malathion and its Metabolites to Fish Poisoning.** Bull Environ Contam Toxicol 16, 283

Cope OB (1965) **Agricultural Chemicals and Freshwater Ecological Systems.** In: Research in Pesticides, 115-128, New York, Academic Press

Cope OB (1966) **Contamination of Freshwater Ecosystem by Pesticides,** J Appl Ecol Suppl 3, 33-44

Courtney W A and Denton G R W (1976) **Persistence of Polychlorinated Biphenyls in the Hard Clam** ***(Mercenaria mercenaria)*** **and Effect upon the Distribution of these Pollutants in the Estuarine Environment,** Environ Pollut 10, 55 - 64

Courtney W A M and Langston W J (1978) **Uptake of Polychlorinated Biphenyl (Aroclor 1254) from Sediment and from Seawater in Two Intertidal Polychaetes,** Environ Pollut 15, 303 - 309

Cowan A A (1981) **Organochlorine Compounds in Mussels from Scottish Coastal Waters,** Environ Pollut Ser B 2, 129 - 143

Cowart RP, Bonner FL and Epps EA Jr (1971) **Rate of Hydrolysis of Seven Organophosphate Pesticides,** Bull Environ Contam Toxicol 6, 231

Cox JL (1971) **DDT Residues in Seawater and Particulate Matter in the California Current System,** Fishery Bull US 69, 443-50

Croll B T (1991) **Pesticides in Surface Waters and Groundwaters,** J Inst Wat Enviro Mangt 5, 389-395

Croll B T (Anglian Water) **The Effects of the Agricultural Use of Herbicides on Fresh Waters,** Paper 13 from WRc

Dabydeen S and Leavitt J R C (1981) **Absorption and Effect of Simazine and Atrazine on** *Elodea canadensis,* Bull Environ Contam Toxicol 26, 381 - 385

Davar P and Wightman JP (1981) in PH Tewari (Ed) **Adsorption from Aqueous Solutions,** Plenum Publ Corp, New York, 163-177

Dave G, Boman A, Johansson-Sjobeck ML and Lidman U (1975), **Metabolism Effects of PCB (Polychlorinated Biphenyls) in the European Eel (*Anguilla anguilla*)**, In: JH Koeman and J JTWA Strick (Eds), Elsevier, Amsterdam pp 197-205

Davies I M and McKie J C (1987) **Accumulation of Total Tin and Tributyltin in Muscle Tissue of Farmed Atlantic Salmon,** Mar Pollut Bull 18 (7), 405 - 407

Davies IM, McKie JC and Paul JD (1986) **Accumulation of Tin and Tributyltin from Antifouling Paints by Cultivated Scallops (*Pecten maximus*) and Pacific Oysters (*Crassostrea gigas*)** Aquaculture, 55, 103-14

Davies IM, Bailey SK and Moore DC (1987) **Tributyltin in Scottish Sea Lochs, as Indicated by Degree of Imposex in the Dogwhelk, *Nucella lapillus*,**Mar Pollut Bull, 18, 400-4

Davies R P and Dobbs A J (1984) **The Prediction of Bioconcentration in Fish,** Wat Res 18, 1253 - 1262

Dawson R and Tiley JE (1977) **Chlorine-Containing Pesticides and Polychlorinated Biphenyls in British Coastal Waters.** Estuar Coastal Mar Sci, 4, 55-69

de Koch W C (1983) **Accumulation of Cadmium and PCBs by *Mytilus edulis* Transplanted from Pristine Water into Pollution Gradients,** Can J Fish Aquat Sci 40 (Suppl 2), 282 - 294

de Koch W C (1986) **Monitoring Bioavailable Marine Contaminants with Mussels (*Mytilus edulis*) in the Netherlands,** Environ Monit Assess 7, 209 - 220

Defoe DL, Veith GD and Carlson RW (1978) **Effects of Aroclor 1248 and 1260 on the Fathead Minnow *Pimephales Promelas*,** J Fish Res Bd Canada, 35, 997-1002

Denton GRW (1974) **The Uptake and Elimination of Poly Chlorinated Biphenyls (PCBs) in the Hardshell Clam *Mercenaria mercenaria*,** PhD thesis London pp 194 1974

Department of Environment (July 1988) **Inputs of Dangerous Substances to Water: Proposals for a Unified System of Control: The Red List**

Department of Environment (1987) **Memorandum Submitted to the House of Commons Environment Committee,** In: Pollution of Rivers and Estuaries, Third Report from the Environment Committee, Session 1986-87, HMSO, London, 1987, Volume II, Appendix 24

Department of Environment, (July 1990) **UK Guidance Note on the Ministerial Declaration,** Third International Conference of the Protection of the North Sea

Department of Environment: **Final Declaration of the Third International Conference on the Protection of the North Sea**

Dewey SL (1986) **Effects of the Herbicide Atrazine on Aquatic Insect Community Structure and Emergence,** Ecology 67, 148-162

Duffey JR and O'Connell (1968) **DDT Residues and Metabolites in Canadian Atlantic Coast Fish,** J Fish Res Bd Canada 25, 189-95

Duinker J C, Hillebrand M T J and Boon J P (1983) **Organochlorines in Benthic Invertebrates and Sediments from the Dutch Wadden Sea; Identification of Individual PCB Components,** Neth J Sea Res 17, 19 - 38

Duursma E K et al (1986) **Partitioning of Organochlorines Between Water, Particulate Matter and Some Organisms in Estuarine and Marine Systems of the Netherlands,** Netherland J Sea Res 20, 239 - 251

Duursma E K, Niewenhuize J and van Liere J M (1989) **Polychlorinated Biphenyl Equilibria in an Estuarine System,** Sci Tot Environ 79, 145 - 155

Ebdon L, Evans K and Hill S (1988) **The Accumulation of Organotins in Adult and Seed Oysters from Selected Estuaries Prior to the Introduction of UK Regulations Governing the Use of Tributyltin-based Antifouling Paints,** Sci Tot Environ 8, 3 (1989), 63 - 84

Ebdon L, Evans K and Hill S (1988) **The Variation of Tributyltin Levels with Time in Selected Estuaries Prior to the Introduction of Regulations Governing the Use of Tributyltin-based Anti-fouling Paints,** Sci Tot Environ, 68, 207 - 223

Edwards C A (c1965) Effects of Pesticies Residues on Soil Invertebrates and Plants

Edwards CA (1973) **Persistent Pesticides in the Environment,** 2nd ed Ohio, CRC Press

EIFAC (1980) **Water Quality Criteria for European Freshwater Fish. Report on Combined Effects on Freshwater Fish and Other Aquatic Life Mixtures of Toxicants in Water.** EIFAC Technical Paper No 37, European Fisheries Advisory Commission, Food and Agriculture Organisation of the United Nations, Rome

Eisler R (1970) **Acute Toxicities of Organochlorine and Organophosphorus Insecticides to Estuarine Fishes,** Tech Pap Bur Sport Fish Wildl (US), 46, 3-12

Elder D L, Fowler S W and Polikarpov G G (1979) **Remobilization of Sediment - Associated PCBs by the Worm,** *Nereis diversicolor*, Bull Environ Contam Toxicol 21, 448 - 452

Ellgehausen H et al (1980) **Factors Determining the Bioaccumulation Potential of Pesticides in the Individual Compartments of Aquatic Food Chains,** Ecotoxicol Environ Safety 4, 134 - 157

Ernst W and Goerke H (1974) **Anreicherung, Verteilung, Umwandlung und Ausscheiclung von DDT C^{14} bei** *Solea solea* **(Pisces: Soleidea),** Mar Biol, 24, 287-304

Ernst W, Foerke H, Eder G and Schaefer RG (1976) **Residues of Chlorinated Hydrocarbons in Marine Organisms in Relation to Size and Ecological Parameters.I PCB, DDT, DDE and DDD in Fishes and Molluscs from the English Channel,** Bull Environ Contam Toxicol 15, 55-65

Ernst W (1977) **Determination of the Bioconcentration Potential of Marine Organisms - A Steady State Approach. 1. Bioconcentration Data for Seven Chlorinated Pesticides in Mussels** *(Mytilus eduiis)* **and their Relationship to Solubility Data,** Chemosphere 6, 731 - 740

Ernst W and Weber K (1978) **Chlorinated Phenols in Selected Estuarine Bottom Fauna,** Chemosphere 11, 867 - 872

Evans C J (1974) **Developments in the Organotin Industry. Tri-organotin Chemicals,** In: Tin Research Institute Publication No 491

Evans DW and Laughlin R (1984) Chemosphere 13, 213-219

Faust SD and Gomma M (1972) **Chemical Hydrolysis of Some Organic Phosphorous and Carbamate Pesticides in Aquatic Environments,** Environ Lett, 3, 171

Feral C and Le Gall S (1982) **Induction Experimentale par un Pollutant Marin (le Tributyletain), de l'activite Neuroendocrine Controlant la Morphogenese du Penis chez les Femelles d'***Ocenebra erinocea* **(Mollusque, Prosobranche Gonochorique),** Compte Rendu Hebdomadaire des Seances de l'Academie de Sciences 295, 627-630

Ferguson DE and Bingham CR (1966) **The Effects of Combinations of Insecticides on Susceptible and Resistant Mosquitofish,** Bull Environ Contam Toxicol 1, 97

Ferrando M D et al (1987) **Acute Toxicity of Organochlorinated Pesticides to the European Eel, *Anguilla anguilla:* the Dependency on Exposure Time and Temperature,** Bull Environ Contam Toxicol 39, 365 - 369

Ferrando M D and Andreu Moliner E (1989) **Effects of Temperature, Exposure Time and Other Water Parameters on the Acute Toxicity of Endosulfan to European Eel, *Anguilla anguilla,*** J Environ Sci and Health Part B, B24, 219 - 224

Foehrenbach J (1972) **Chlorinated Pesticides in Estuarine Organisms,** J Wat Pollut Control Fed 11, 619 - 624

Fowler S W, Polikarpov G G, Elder D L, Parsi P and Villeneuve J P (1978) **Polychlorinated Biphenyls : Accumulation from Contaminated Sediments and Water by the Polychaete *Nereis diversicolor,*** Mar Biol 48, 303 - 309

Frank R, Armstrong AE, Boelens RG, Braun HE and Douglas CW (1974) **Organochlorine Insecticide Residues in Sediment and Fish Tissues, Ontario, Canada** Pestic Monit J 7, 165-80

Freed V H and Chiou C T (1982) **Physiochemical Factors in Routes and Rates of Human Exposure to Chemicals** In: Environmental Health Chemistry Chap 3 (ed J C McKinney) pp 59 - 74 Ann Arbor Science, Ann Arbor

Friends of the Earth (1991) **Poisonous Fish**

Frisque GE, Galoux M and Bernes A (1983) **Accumulation de deux Micropollutants (les Polychlorobiphenyles et le gamma-HCH) par des bryophytes aquatiques de la Meuse,** Meded Fac Landbouwwet Rijksuniv Gent, 48/4, 971-983

Gallissot And Mouvet, in prep (1985)

Galtstoff PS (1964) **The Americal Oyster** *Crassostrea virginica* **Gmelin,** Fish Bull, US Fish Wildl Serv 64, 1-480

Gannon JE and Beeton AM (1971) J Water Pollut Control Fed 43, 392

Gault NFS, Tolland ELC and Parker JG (1983) **Spatial and Temporal Trends in Heavy Metal Concentrations in Mussels from Northern Ireland Coastal Waters,** Mar Biol 77, 307-316

Geyer H, Sheehan P, Kotzias D, Freitag D and Korte F (1982) **Prediction of Ecotoxocological Behaviour of Chemicals : Relationship between Physicochemical Properties and Bioaccumulation of Organic Chemicals in the Mussel, *Mytilus edulis,*** Chemosphere 11, 1121 - 1134

Gibbs P E, Bryan G W, Pascoe P L and Burt G R (1987) **The Use of the Dogwhelk Nucella lapillus, as an Indicator of Tributyltin (TBT) Contamination,** J Mar Biol Ass UK, 67, 507 - 523

Giese AC (1969) **A New Approach to the Biochemical Composition of the Mollusc Body,** Oceanogr Mar Biol Ann Rev 7, 175-229

Gobas F A P C, Opperhuizen A and Hutzinger O (1986) **Bioconcentraion of Hydrophobic Chemicals in Fish: Relationship with Membrane Permeation,** Environ Toxicol Chem 5, 637 - 646

Gobas F A P C, McNeil E J, Lovett-Doust L and Haffner G D (1991) **Bioaccumulation of Chlorinated Aromatic Hydrocarbons in Aquatic Macrophytes,** Environ Sci Technol 25 (5), 924 - 929

Goerke H and Ernst W (1977) **Fate of ^{14}C-labelled di- tri-, and Pentachloro Biphenyl in the Marine Annelid *Nereis virens,* I Accumulation and Elimination After Oral Administration,** Chemosphere 6, 551-558

Goerke H (1979) ***Nereis virens* in Marine Pollution research: Culture Methods and Oral Administration of a Polychlorinated Biphenyl,** Veröff Inst Meeresforsch Bremerh 17, 151-161

Goerke H (1984b) **Temperature - Dependent Elimination of 2, 4, 6, 2', 4' - pentachloro [U-^{14}C] Biphenyl in *Nereis virens* (Polychaeta)** Arch Environ Contam Toxicol 13, 347-355

Goerke H and Ernest W (1986) **Elimination of Pentachlorobiphenyls by** *Nereis virens* **(Polychaeta) in the Laboratory and the Marine Environment** Chem Ecol 2, 263 - 285

Goerke H and Weber K (1990) **Population Dependent Elimination of Various Polychlorinated Biphenyls in** *Nereis diversicolor* **(Polychaeta),** Mar Environ Res 29, 205 - 226

Goldberg ED (1975) **The Mussel Watch - A First Step in Global Marine Monitoring,** Mar Pollut Bull 6, 111

Goldberg ED, Bowen VT, Farrington JW, Harvey G, Martin JH, Parker PL, Risebrough RW, Robertson W, Schneider E and Gamble E (1978) **The Mussel Watch,** Env Conserv, 5, 101-25

Goldberg E D (1986) **The Mussel Watch Concept,** Environ Monit Assess 7, 91 -103

Gomme J W and Shurvell S et al (1991) **Hydrology of Pesticides in a Chalk Catchment : Surface Waters,** J IWEM 5 Oct, 546 - 552

Gosset R W, Brown D A and Young D R (1983) **Predicting the Bioaccumuation of Organic Compounds in Marine Organisms using Octanol-Water Partition Coefficients,** Mar Pollut Bull 14, 387 -392

Goulding K H and Wynne-Ellis S (1981) **The Interaction of DDT with Two Species of Freshwater Algae,** Environ Pollut Ser A 25, 271 - 290

Grant B F (1976) **Endrin Toxicity and Distribution in Freshwater: A Review,** Bull Environ Contam Toxicol 15 (3), 283 - 290

Grauby A, Foulquier C, Descamps B and Jaulent Y (1973) **Résultats relatifs à la toxicité d'effluents industriels de la région de Fos et de l'étang de Berre sur les Anguilles, les daphnies, les loups et les muges,** CEA, Labo, Radio écologie continentale, contract No VEN - 0368, Agence de Bassin Rhône Méditerranée-Corse, pp 135

Green et al (1986) **Determination of Chlorinated Hydrocarbons in Coastal Waters Using a Moored in situ Sampler and Transplanted Live Mussels,** Mar Pollut Bull 17, 324 - 329

Griesbach S et al (1982) **An Allometric Model for Pesticide Bioaccumulation,** Can J Fish Aquat Sci 39, 727 - 735

Guiney PD, Melancon MJ, Lech JJ and Peterson RW (1979) **Effects of Egg and Sperm Maturation and Spawning on the Distribution and Elimination of Polychlorinated Biphenyl in Rainbow Trout** (*Salmo gairdneri*), Toxicol Appl Pharmacol 47, 261-272

Gunkel G (1979a) **Aufnahme und Anreicherung van Schadstoffen in Einem Aquatischen Nahrungskettengefuge,** Arbeitsberichte Okologie / Umwelltechnik 31 - 45, Konstanz

Gunkel G and Streit B (1980) **Mechanisms of Bioaccumulation of a Herbicide (atrazine, s-triazine) in a Freshwater Mollusc** (*Ancylus fluviatilis* **Müll) and a Fish** (*Ceregonus fera* **Jurnine**), Wat Res 14, 1573 - 1584

Gunkel G (1981) **Bioaccumulation of a Herbicide (Atrazine, S-triazine) in a Whitefish** (*Coregonus fera* **J**)**: Uptake and Distribution of the Residue in Fish,** Arch Hydrobiol Suppl 59, 252 - 287

Gunther FA, Westlake WE and Jaglan PS (1968) **Reported Solubilities of 738 Pesticide Chemicals in Water,** Residue Rev, 20, 1-148

Gustafson CG (1970) PCBs - **Prevalent and Persistent,** Environ Sci and Technol 4, 814-819

Hamelink JL, Waybrant RC and Ball RC (1971) **A Proposal: Exchange Equilibria Control the Degree Chlorinated Hydrocarbons are Biologically Magnified in Lentic Environments,** Trans Am Fish Soc 100, 207-214

Hamelink JL and Waybrant RC (1976) **DDE and Lindane in a Large-Scale Model Lentic Ecosystem,** Trans Amer Fish Soc 105, 124-34

Hamilton RM (1985) **Discharge of Pesticides to the Rivers Mole and Taw, Their Accumulation in Fish Flesh and Possible Effects on Fish Stocks,** J Fish Biol 27 (Suppl A), 139-149

Hamilton E I (1989) **Chemical Contamination of French Coasts - The Results of a Ten Year Mussel Watch,** Mar Pollut Bull 20, 523 - 528

Hansen P-D (1980) **Uptake and Transfer of the Chlorinated Hydrocarbon Lindane (gamma BHC) in a Laboratory Freshwater Food Chain,** Environ, Pollut (Series A) 21, 97 - 108

Hansen DJ and Wilson AJ (1970) **Significance of DDT Residues from the Estuary near Pensacola, Fla,** Pestic Monit J 4, 51-6

Harding GCH and Vass WP (1978) **Uptake from Seawater of DDT by Marine Planktonic Crustacea,** J Fish Res Bd Can 36, 247-254

Hargrave BT and Phillips GA 91976) **DDT Residues in Benthic Invertebrates and Demersal Fish in St Margaret's Bay, Nova Scotia,** J Fish Res Bd Canada 33, 1692-8

Hattula ML, Janatuinen J, Särkkä J and Paasivirta J (1978a) **A Five-year Monitoring Study of the Chlorinated Hydrocarbons in the Fish of a Finnish Lake Ecosystem,** Environ Pollut 15, 121-39

Haug A, Melsom S and Omang A (1974) **Estimation of Heavy Metal Pollution in Two Norwegian Fjord Areas by Analysis of the Brown Alga** ***Ascophyllum nodosum,*** Environ Pollut 7, 173-93

Hawker D W and Connell D W (1986) **Bioconcentration of Lipophilic Compounds by Some Aquatic Organisms,** Ecotoxicol and Environ Safety 11, 184 - 197

Hawker D W and Connell D W (1989) **Factors Affecting Bioconcentration of Trace Organic Contamination in Waters,** Wat Sci Technol 21, 147 - 150

x Hellawell JM (1986) **Biological Indicators of Freshwater Pollution and Environmental Management,** Elsevier Applied Science, London

Henderson C, Johnson WL and Inglis I (1969) **Organochlorine Insecticide Residues in Fish, National Pesticide Monitoring Program,** Pestic Monit J 3, 145-71

Henderson C, Inglis I and Johnson L (1971) **Organochlorine Insecticide Residues in Fish, Fall 1969 National Pesticide Monitoring Program,** Pestic Monit J 5, 1-11

Henderson RJ and Tocher DR (1987) **The Lipid Composition and Biochemistry of Freshwater Fish,** Progress in Lipid Research 26, 281-347

Herrmann SJ (1970a) **Systematics, Distribution and Ecology of Colorado Hirudinea,** Amer Midl Natur 83, 1-37

Herve S, Heinonen P, Paukku R, Knuutila M, Koistinen J and Paasivirta J (1988) **Mussel Incubation Method for Monitoring Organochlorine Pollutants In Water Courses - 4 Year Application in Finland,** Chemosphere 17 (10), 1945 - 1961

Hesig-Gunkel G and Gunkel G (1981) **Distribution of a herbicide (atrazine, s-triazine) in** ***Daphnia pulicaria*****: a New Approach for Determination,** Arch Hydrobiol / Supp (im Druck)

Hider RC, Mason CF and Bakaj ME (1982) **Chlorinated Hydrocarbon Pesticides and Polychlorinated Biphenyls in Freshwater Fishes in the United Kingdom 1980-1981,** The Vincent Wildlife Trust, London

Hofmann A and Winkler S (1990) **Effects of Atrazine in Environmentally Relevant Concentration on Submerged Macrophytes**, Arch Hydrobiol 118, 69 - 79

Holden AV (1962) **A Study of the Absorbtion of C^{14}labelled DDT from Water by Fish**, Ann Appl Biol 50, 467-77

Holden AV (1972b) **The Effects of Pesticides on Life in Fresh Waters**, Proc R Soc Lond B, 180, 383-394

Holden AV (1973) **Effects of Pesticides on Fish**, In: Environmental Pollution by Pesticides, (CA Edwards, Ed) pp 213-53 London, Plenum Press

Holdgate MW (Ed) (1971) **The Seabird Wreck of 1969 in the Irish Sea.** NERC Publ Series C, No 4

Howard LS and Brown BE (1983) **Natural Variations in Tissue Concentration of Copper, Zinc and Iron in the Polychaete *Nereis diversicolor***, Mar Biol 78, 87-97

Huckins J N, Petty J D and England D C (1986) **Distribution and Impact of Trifluralin, Atrazine and Fonofos Residues in Microcosms Stimulating a Northern Prairie Wetland**, Chemosphere Vol 15, No 5, 563 - 588

Hummel H, Bogaards R H, Nieuwenhuize J et al (1990) **Spatial and Seasonal Differences in the PCB Content of the Mussel**, *Mytilyus edulis*, Sci Tot Environ 92, 155 - 163

Huschenbeth E (1977) **Uberwachung der Speicherung von chlorierten Kohlenwasserstoffen im Fisch**, Arch Fischerei Wiss 28 (2/3), 173-186

Hutzinger O, Van Lelyvland IH and Zoeteman BCJ (1978) **Aquatic Pollutants: Transformation and Biological Effects**, Pergamon Press, London

Isnard P and Lambert S (1988) **Estimating Bioconcentration Factors from Octanol-Water Partition Coefficient and Aqueous Solubility**, Chemosphere 17 (1), 21 - 34

James MO, Fouts JR and Bend JR (1977) **Xenobiotic Metabolizing Enzymes in Marine Fish**, In Pesticides in Aquatic Environments (Ed Khan MAQ) pp 171-189 Plenum Press, New York

Jangaard PM, Brockerhoff H, Burgher RD and Hoyle RJ (1967a) **Seasonal Changes in General Condition and Lipid Content of Cod from Inshore Waters**, J Fish Res Bd Canada, 24, 607-12

Jangaard PM, Ackman RG and Sipos JC (1967b) **Seasonal Changes in Fatty Acid Composition of Cod Liver, Flesh, Roe and Milt Lipids**, J Fish Res Bd Canada 24, 613-27

Jarvinen AW, Hoffman MJ and Thorslund TW (1977) **Long-term Toxic Effects of DDT Food and Water Exposure on Fathead Minnows, (*Pimpephales promelas*)**, J Fish Res Bd Canada 34, 2089-103

Jensen S, Johnels AG, Odsjö T, Olsson M and Olterlind G (1970), **PCB - Occurrence in Swedish Wildlife,** In: Proceedings of the PCB Conference, Wenner-Gren Centre, Stockholm, 29 Sept 1970, Available from the Research Secretariat, Statens Naturvardsverk, Fack, 171 20 Solna, Sweden

Jensen S, Johnels AG, Olsson M and Olterlind G (1972a) **DDT and PCB in Herring and Cod from the Baltic, the Kattegat and the Skagerrak,** Ambio Spec Rep No 1, 71-85

Jensen S, Olsson M and Vaz R (1975) **Levels of DDT and PCB in Littoral Fishes along the Swedish Coast,** In: Proceedings of Soviet-Swedish Symposium on the Baltic Pollution, 3rd, Stockholm, Sweden, September 1975

Johnson DW (1968) **Pesticides and Fishes - A Review of Selected Literature,** Trans Am Fish Soc 97, 398-424

Johnson DW (1973) **Pesticide Residues in Fish,** In: Environmental Pollution by Pesticides, (CA Edwards, Ed) pp 181-212 London, Plenum Press

Johnson B T, Saunders C R, Sanders H O and Campbell R S (1971) **Biological Magnification and Degradation of DDT and Aldrin by Freshwater Invertebrates,** J Fish Res Bd Can 28, 705 - 709

x Jurgensen T A and Hoagland K D (1990) **Effects of Short-term Pulses of Atrazine on Attached Algal Communities in a Small Stream,** Arch Environ Contam Toxicol 19, 617 - 623

Kammann U, Knickmeyer R and Steinhart H (1990) **Distribution of Polychlorobiphenyls and Hexachlorobenzene in Different Tissues of the Dab (*Limanda limanda*) in Relation to Lipid Polarity,** Bull Environ Contam Toxicol 45 (4), 552 - 559

Kanazawa J (1981) **Measurement of the Bioconcentration Factors of Pesticides by Freshwater Fish and their Correlation with Physiochemical Properties of Acute Toxicities,** Pestic Sci 12, 417 - 424

Karickhoff SW, Brown DS and Scott TA (1979) Water Res 13, 241

Katz M and Chadwick GG (1961) **Toxicity of Endrin to Some Pacific Northwest Fishes,** Trans Am Fish Soc 90, 394-397

Kawamoto K and Urano K (1990) **Parameters for Predicting Fate of Organochlorine Pesticides in the Environment 3. Biodegradation Rate Constants,** Chemosphere 21 (10 -1), 1141 - 1152

Kelso JRM, Maccrimmon HR and Ecobichon 91970) **Seasonal Insecticide Residue Changes in Tissues of Fish from the Grand River, Ontario,** Trans Amer Fish Soc 99, 423-6

Kemp MV and Wightman JP (1981) Virginia J Sci 32, 34

Kenezovich J P, Harrison FL and Wilhelm R G (1987) **The Bioavailability of Sediment - Sorbed Organic Chemicals : A Review,** Water, Air, Soil Pollution 32, 233 - 245

Kerr SR and Vas WP (1973) **Pesticide Residues in Aquatic Invertebrates,** In: Environmetnal Pollution by Pesticides (ed CA Edwards) pp 134-180, London and New York, Plenum Press

Kettle WD, DeNoyelles F, Heacock BD and Kadoum AM (1987) **Diet and Reproductive Success of Blue Gill Recovered from Experimental Ponds Treated with Atrazine,** Bull Environ Contam Toxicol 38, 47-52

Kinter WB, Merkens LS, Janicki RH and Guarino AM (1972) **Studies on the Mechanism of Toxicity to DDT and Polychlorinated Biphenyls (PCBs): Disruption of Osmoregulation in Marine Fish,** Environmental Health Perspectives, US Dept of Health, Education and Welfare, NIH 88146 pp 169-173

Knickmeyer R and Steinhart H (1989) **Cyclic Organochlorines in the Whelks** *Bussinum undatum* **and** *Neptunea antiqua* **from the North Sea and the Irish Sea,** Mar Pollut Bull 20, 433 - 437

Knickmeyer R and Steinhart H (1989) **On the Distribution of Polychlorinated Biphenyl Congeners and Hexachlorobenzene in Different Tissues of Dab (***Limanda limanda***) from the North Sea.** Chemosphere 19 (8 - 9), 1309 - 1320

Knox GA (1977) **The Role of Polychaetes in Benthic Soft-bottom Communities,** In: Reish DJ and Fauchald K (Eds) Essays on Polychaetous Annelids in Memory of Dr Olga Hartman, pp 547-604, Allan Hancock Foundation, Univ of So California, Los Angeles, pp 604

Kobayashi K (1978) **Metabolism of Pentachlorophenol in Fishes.** In Pentachlorophenol: Chemistry, Pharmacology and Environmental Toxicology, (ed) Rao K R, Environmental Science Research No 12, 1978, Plenum Press, New York

Kovats Z E and Ciborowski J J H (1989) **Aquatic Insect Adults as Indicators of Organochlorine Contamination,** J Great Lakes Res 15, 623 - 634

Kruse VR, Boek K and Wolf M (1983) **Der gehalt an organochlor-pestiziden und polychlorierten biphenylen in Elbeaalen,** Archiv für Lebensmittelhygiene, 34, 81-86

Kupfer D (1967) **Effects of Some Pesticides and Related Compounds on Steroid Function and Metabolism,** Residue Rev 19, 11-30

Langston W J (1978a). **Accumulation of Polychlorinated Biphenyls in the Cockle** *Cerastoderma edule* **and the Tellis** *Macoma balthica,* Mar Biol 45, 265 - 272

Langston W J (1978b) **Persistence of Polychlorinated Biphenyls in Marine Bivalves,** Mar Biol 46, 35 - 40

Langston W J, Burt G R and Mingjlang Z (1987) **Tin and Organotin in Water, Sediments and Benthic Organisms of Poole Harbour,** Mar Pollut Bull 18, 634 - 639

Langston W J, Bryan G W, Burt G R and Gibbs P E (1990) **Assessing the Impact of Tin and TBT in Estuaries and Coastal Regions,** Functional Ecol 4, 433 - 443

Larsson P (1983) **Transport of ^{14}C-labelled PCB Compounds from Sediment to Water and from Water to Air in Laboratory Model Systems,** Water Res 17, 1317-26

Larsson P (1984) **Uptake of Sediment Released PCBs by the Eel *Anguilla anguilla* in Static Model Systems,** Ecol Bull 36, 62-67

Larsson P (1984) **Transport of PCBs from Aquatic to Terrestrial Environments by Emerging Chironomids,** Environ Pollut 34, 283 - 289

Larsson P, Hamrin S and Okla L (1991) **Factors Determining the Uptake of Persistent Pollutants in an Eel Population (*Anguilla anguilla*)** Environ Pollut 69 (1), 39 - 50

Laughlin R B Jr and Linden O (1985) **Fate and Effects of Organotin Compounds,** Ambio 14, 88 - 94

Laughlin R B Jr and Linden O (1987) **Tributyltin - Contemporary Environmetal Issues,** Ambio 16, 252 - 256

Laughlin R B Jr, French W and Guard H E (1986) **Accumulation of Bis (Tributyltin) Oxide by the Marine Mussel,** *Mytilus edulis,* Environ Sci Technol 20, 844 - 890

Lech JJ and Bend JR (1980) **Relationship Between Biotransformation and the Toxicity and Fate of Xenobiotic Chemicals,** Enviro Hlth Persp 34, 115-131

Lee RF (1986) **Metabolism of Bis (tributyltin) Oxide by Estuarine Organisms.** In Oceans 86 Conference Record, Vol 4 Organotin Symposium, 1182-1188, New York Institute of Electrical and Electronics Engineers

Lenon HL (1968) **Translocations and Storage Equilibria for Minnows Involving Sublethal Levels of Dieldrin in Aquatic Ecosystems,** PhD thesis, Michigan State University, Michigan, pp 85

Lichtenstein EP and Schulz KR (1964) **The Effects of Moisture and Micro organisms on the Persistence and Metabolism of some Organophosphorus Insecticides in Soils with Special Emphasis on Parathion,** J Econ Entomol, 57, 618

Linko RR, Kaitaranta J, Rantamäki P and Eronen L (1974) **Occurrence of DDT and PCB Compounds in Baltic Herring and Pike from the Turku Archipelago,** Environ Pollut 7, 193-207

Linton LR, Davies RW and Wrona FJ (1983a) **The Effects of Water Temperature, Ionic Content and Total Dissolved Solids on *Nephelopsis obscura* and *Erobdella punctata* (Hirudinoidea, Erpobdellidae) 1 Mortality,** Holatct Ecol 6, 59-63

Livingston R J (1977) **Review of Current Literature Concerning the Acute and Chronic Effects of Pesticides on Aquatic Organisms,** CRC Critical Reviews in Environmental Control 7, 325, 351

Livingston RJ, Thompson MP and Meeter DA (1978) **Long-term Variation of Organochlorine Residues and Assemblages of Epibenthic Organisms in A Shallow North Florida (USA) Estuary,** Mar Biol 46, 355-72

Lopez E, Peignoux-Deville J, Lallier F, Martelly E and Fontaine YA (1981a) **Anguilles Contaminées par les Hydrocarbures après l'échouage de l'Amoco-Cadiz, Modifications Histopathologiques des Ovaires, des Branchies et de Glandes Endocrines,** CR Acad Sci Paris, 292 407-411

Lopez E, Leloup-Hatey J, Hardy A, Lallier F, Martelly E, Oudot J, Peignoux-Deville J and Fontained YA (1981b) **Modifications Histopathologiques et stress chez des Anguilles Soumises à une Exposition Prolongée aux Hydrocarbures,** In: Amoco-Cadiz; Conséquences d'une Pollution Accidentelle par Hydrocarbures, CNEXO, Paris pp 645-653

Lord KA, Briggs GG, Neale MC and Manlove R (1980), Pestic Sci, 11, 401

Lovern JA (1938) **Fat Metabolism in Fishes XIII: Factors Influencing the Composition of Depot Fat of Fishes,** Biochem J 32, 1214-1224

Lu P-Y, Metcalfe R L and Cole L (1978) **The Environmental Fate of 14 C-pentachlorophenol in Laboratory Model Ecosystems,** in : Pentachlorophenol : Chemistry, Pharmacology and Environmental Toxicology (ed) Rao K R, Environmental Science Research No 12, 1978, Plenum Press, New York

Macek KJ and Korn S (1970) **Significance of the Food Chain in DDT Accumulation by Fish,** J Fish Res Bd Can, 27, 1496-8

Macek KJ, Rodgers CR, Stalling DL and Korn S (1970) **The Uptake, Distribution and Elimination of Dietary ^{14}C-DDT and ^{14}C-Dieldrin in Rainbow Trout,** Trans Am Fish Soc, 99, 689-95

Macek KJ, Buxton KS, Sauter S, Gnilka S and Dean JW (1976) **Chronic Toxicity of Atrazine to Selected Aquatic Invertebrates and Fishes,** US EPA Rep 600/3-76-047, pp 58

MacKay D (1982) **Correlation of Bioconcentration Factors,** Environ Sci Technol, 16, 274-8

MAFF **Report of the Working Party on Pesticide Residues: 1985 - 1988, (Freshwater Eels)**

Maguire RJ, Wong PTS and Rhamey JS (1984) **Accumulation and Metabolism of tri-n-butyltin Cation by a Green Algae, *Ankistrodesmus fakatus*,** Can J Fish Aquat Sci 41, 537-540

Maki, AW (1979) **Aquatic Test Organisms and Methods useful for Assessment of Chronic Toxicitiy of Chemical,** In Analyzing the Hazard

Evaluation Process, (KL Dickson, AW Maki and J Cairns, Jr, Eds), 83-100, Water Quality Sections - American Fisheries Society, Washington DC

Marchand M (1981) **Amoco Cadiz: Bilan du Colloque sur les Conséquences d'une Pollution Accidentelle par les Hydrocarbures,** Rapp Sci Tech Publ CNEXO, Paris 44, pp 86

Marcus J M and Renfrow R T (1990) **Pesticides and PCBs in South Carolina Estuaries,** Mar Pollut Bull 21, 96 - 99

Markwell R D, Connell DW, Gabric A J (1989) **Bioaccumulation of Lipophilic Compounds from Sediments by Oligochaetes,** Water Res 23, 1443 - 1450

Martin M (1985) **State Mussel Watch: Toxics Surveillance in California,** Mar Pollut Bull 16, No 4

Mauck W L, Mayer FL and Holz D D (1976) **Simazine Residue Dynamics in Small Ponds,** Bull Environ Contam Toxicol Vol 16, No 1, 1 - 8.

Mauck WL, Mehrle PM and Mayer FL (1978) **Effects of the Polychlorinated Biphenyl Archlor 1254 on Growth, Survival and Bone Development in Brook Trout** *(Salvelinus fontinalis)* J Fish Res Board Can 35, 1084-8

Mayer FL, Street JC and Neuhold JM (1970) **Organochlorine Insecticide Interactions Affecting Residue Storage in Rainbow Trout,** Bull Environ, Contam Toxicol, 5,300-310

McKie J C (1987) **Determination of Total Tin and Tributyltin in Marine Biological Materials by Electrothermal Atomic Absorption Spectrometry,** Anal Chim Acta 197, 303 - 308

McLeese D W, Metcalfe C D and Pezzak D S (1980) **Uptake of PCBs from Sediment by** *Nereis virens* **and** *Crangon septemspinosa,* Arch Environ Contam and Toxicol 9, 507 - 518

Meeks RK (1968) **The Accumulation of ^{36}Cl Ring-labelled DDT in a Freshwater Marsh,** J Wildl Mgmt, 32, 376-398

Meeks RL and Peterle TJ (1967) **RF Project 1974, Report No C00-1358-3** The Ohio State University, Res Found Columbus

Mellanby K (1970) **Pesticides and Pollution,** New Naturalist Series, London, Collins

Metcalf RL (1977) **Model Ecosystem Studies of Bioconcentration and Biodegradation of Pesticides,** In: Pesticides in Aquatic Environments (Ed MAQ Khan) pp 127-144 New York, Plenum Press

Metcalfe J L, Fox M E, Carey J H (1984) **Aquatic Leeches (Hirudinea) as Bioindicators of Organic Chemical Contaminants in Freshwater Ecosystems,** Chemosphere 13 (1), 143 - 150

Metcalfe J L, Fox ME and Carey J H (1988) **Freshwater Leeches (Hirudinea) as a Screening Tool for Detecting Organic Contaminants in the Environment**, Environ Monit Assess 11, 147 - 169

Metcalfe J L and Hayton A (1989) **Comparison of Leeches and Mussels as Biomonitors for Chlorophenol Pollution**, J Great Lakes Res 15, 654 - 668

Minchin D, Duggan C B and King W (1987) **Possible Effects of Organotins on Scallop Recruitment**, Mar Pollut Bull 18, 604 - 608

Ministry of Agriculture, Fisheries and Food (1990) **Directorate of Fisheries Research, Aquatic Environment Monitoring Report No 22: Monitoring and Surveillance of Non-Radioactive Contaminants in the Aquatic Environment 1984-1987**, Lowestoft

Miossec L and Bocquene G (1986) **Toxicité Aiguë et Effects Sublétaux, Après une Courte Exposition, de Différents Nonylphénol Polyéthoxylés sur la Civelle**, Rev Trav Inst Pêches Marit 48 (1-2), 77-84

Mohlenberg F and Kiorboe T (1983) Mar Pollut Bull 14, 57-60

Moriarty F (1988) **Ecotoxicology, The Study of Pollutants In Ecosystems**, Academic Press Ltd, London

Morrison B R S and Wells D E (1981) **The Fate of Fenitrothion in a Stream Environment and its Effect on the Fauna, Following Aerial Spraying of a Scottish Forest**, Sci Total Environ 19, 233 - 252

Mosser JL, Teng TC, Walter WG and Wurster CF (1974) **Interactions of PCBs, DDT and DDE in a Marine Diatom**, Bull Environ Contam Toxicol 12, 665

Mount DI (1962) **Chronic Effects of Endrin on Bluntnose Minnows and Guppies**, US Fish Wildl Serv Res Rep 58, 1-38

Mouvet C (1985) **The Use of Aquatic Bryophytes to Monitor Heavy Metal Pollution of Freshwaters as Illustrated by Case Studies**, Verh Internat Verein Limnol, 22, 2420-2425

Mouvet C, Galoux M and Bernes A (1985) **Monitoring of Polychlorinated Biphenyls (PCBs) and Hexachlorocyclohexanes (HCH) in Freshwater Using the Aquatic Moss Cinclidotus danubicus**, Sci Total Environ 44, 253 - 267

Mouvet C, Gallissot B, Wagner P and Ollivier M (1987) **Micropollutants Organochlores dans le Bassin Rhin-Meuse Rapport de Contrat à l'Agence Financière de Bassin Rhin-Meuse, F-57160**, Moulins-les-Metz, France

Muir D C G, Rawn G P, Townsend B E, Lockhart W L and Greenhalgh R (1985) **Bioconcentration of Cypermethrin, Deltamethrin, Fenvalerate and Permethrin by *Chironomus tentans* Larvae in Sediment and Water**, Environ Toxicol Chem 4, 51 - 61

Muirhead-Thompson RC (1971) **Pesticides and Freshwater Fauna**, Academic Press, London and New York

Murphy PG (1970) **Effects of Salinity on Uptake of DDT, DDE and DDD by Fish**, Bull Environ Contam Toxicol, 5, 404-407

Murphy PG and Murphy JV (1971) **Correlations Between Respiration and Direct Uptake of DDT in the Mosquito Fish *Gambusia affinis***, Bull Environ Contam Toxicol 6, 144-155

Murray A J (1982) **Trace Metals and Organochlorine Pesticide and PCB Residues in Mussels for England and Wales, 1978**, Chem Ecol 1, 33 - 45

Murty AS (1986) **Toxicity of Pesticides to Fish**, Volume 1 CRC Press, Boca Raton, Florida, pp 178

Nakamura T and Mochida K (1988) **Kinetic Evaluation of the Uptake and Decomposition of Pesticides by Algae**, J Pestic Sci 13 (1), 93 - 98

Neff JM (1979) **Polycyclic Aromatic Hydrocarbons in the Aquatic Environment**, Applied Science, New York

Neff JM, Cox BA, Dixit D and Anderson JW (1976) **Accumulation and Release of Petroleum-derived Aromatic Hydrocarbons by Four Species of Marine Animals**, Mar Biol 38, 279-289

Nelson A and Donkin P (1985) **Processes of Bioaccumulation : The Importance of Chemical Speciation**, Mar Pollut Bull Vol 16, No 4, 164 - 169

Neuhoff H-G (1979) **Influence of Temperature and Salinity on Food Conversion and Growth of Different Nereis Species (Polychaeta, Annelida)**, Mar Ecol Prog Ser 1, 255-262

Niagra River Toxics Committee (1984) **Joint Publication of Environmental Protection Agency, New York Department of Environmental Conservation,** Minister of Environment (Ontario), Environment Canada

Niimi A J (1991) **The Solubility of Organic Chemicals in Octanol, Triolein, and Cod Liver Oil and Relationships Between Solubility and Partition Coefficients**, Wat Res Vol 25, No 12, 1515 - 1521

Nimmo DR, Hansen DJ, Couch JA, Cooley NR, Parrish PR and Lowe JI (1975), **Toxicity of Aroclor 1254 and Its Physiological Activity in Several Estuarine Organisms**, Archs Envir Contam Toxicol 3, 22-29

NRA South-west Region (March 1990) EC **Dangerous Substances Directive Monitoring 1989: Report of Environmental Protection Manager**

NRA (Brian Barnett - Anglian Region) (May 1990) **Draft Guidelines for a Bio-accumulation Programme for the Monitoring of Persistent Contaminants in Estuaries and Coastal Waters.**

NRA (Jan 1991) **Final Version: NRA Baseline Estuary and Coastal Water Monitoring Programme**

Ofstad E B and Martinsen K (1983) **Persistent Organochlorine Compounds in Seals from Norwegian Coastal Waters**, Ambio Vol 1 p?

Oliver B G (1984) **Uptake of Chlorinated Organics from Anthropogenically Contaminated Sediments by Oligochaete Worms,** Can J Fish Aquat Sci 41, 878 - 883

Oliver BG (1987) **Biouptake of Chlorinated Hydrocarbons from Laboratory-spiked and Field Sediments by Oligochaete Worms,** Environ Sci Technol 21, 785-790

Olsson M and Jensen S (1978) **Seasonal Variation of PCB Levels in Fish - An Important Factor in Planning Aquatic Monitoring Programs,** Ambio 7, 66 - 69

Olsson M and Reutergardh L (1986) **DDT and PCB Pollution Trends in the Swedish Aquatic Environment,** Ambio 15, 103 - 109

Opperhuizen A and Stokkel R C A M (1988) **Influence of Contaminated Particles on the Bioaccumulation of Hydrophobic Organic Micropollutants In Fish,** Environ Pollut 15 (3), 165 - 177

Paasivirta J et al (1983) **Food Chain Enrichment of Organochlorine Compounds and Mercury in Clean and Polluted Lakes in Finland,** Chemosphere 12, 239 - 252

Paasivirta J (1988) **Organochlorine Compounds in the Environment,** Wat Sci Technol 20, 119 - 129

Page D S (1989) **An Analytical Method for Butyltin Species in Shellfish,** Mar Pollut Bull Vol 20, No 3, 129 - 133

Parker JG and Wilson F (1975) **Incidence of Polychlorinated Biphenyls in Clyde Seaweed,** Mar Pollut Bull 6 (3), 46-47

Pavlou S P and Dexter R N (1979) **Distribution of PCBs in Estuarine Ecosystems. Testing the Concept of Equilibrium Partitioning in the Marine Environment,** Environ Sci Technol 13 (1) 65 - 70

Peakall DB (1967) **Pesticide-Induced Enzyme Breakdown of Steroids in Birds,** Nature Lond 216, 505-506

Pelizzetti E, Maurino V, Minero C, Carlin V, Pramauro E, Zerbinati O and Tosato M (1990) **Photocatalytic Degradation of Atrazine and Other s-triazine Herbicides,** Environ Sci Technol 24, 1559 - 1565

Phillips DJH (1976a) **The Common Mussel *Mytilus edulis* as an Indicator of Pollution by Zinc, Cadmium, Lead and Copper: I Effects of Environmental Variables on Uptake of Metals,** Mar Biol 38, 59-69

Phillips DJH (1976b) **The Common Mussel *Mytilus edulis* as an Indicator of Pollution by Zinc, Cadmium, Lead and Copper: II Relationship of Metals in the Mussel to those Discharged by Industry,** Mar Biol 38, 71-80

Phillips DJH (1977) **The Use of Biological Indicator Organisms to Monitor Trace Metal Pollution in Marine and Estuarine Environments - A Review,** Environ Poll, 13, 281-317

Philips D J H (1978) **Use of Biological Indicator Organisms to Quantitate Organochlorine Pollutants in Aquatic Environments - A Review,** Environ Pollut 6, 167 - 229

Phillips DJH (1979a) **Trace Metals in the Common Mussel, *Mytilus edulis* (L) and in the Alga *Fucus vesiculosus* (L) from the Region of the Sound (Oresund),** Environ Pollut 18, 31-43

Phillips DJH (1980) **Quantitative Aquatic Biological Indicators: Their Use to Monitor Trace Metal and Organochlorine Pollution,** Applied Science Publishers, Barking, Essex, England

Phillips D J H (1985) **Organochlorines and Trace Metals in Green-lipped Mussels (*Perna viridis*) from Hong Kong Water: a Test of Indicator Ability,** Mar Biol Prog Ser 21, 251 - 258

Philips D J H and Segar D A (1986) **Use of Bioindicators in Monitoring Conservative Contaminants : Programme Design Imperatives,** Mar Pollut Bull 17, 10 - 17

Pickering QH, Henderson C and Lemke AF (1962) **The Toxicity of Organic Phosphorus Insecticides to Different Species of Warm Water Fishes,** Trans Amer Fish Soc 91, 175-84

Pierce R H and Victor D M (1978) **The Fate of Pentachlorophenol in an Aquatic Ecosystem** In: Pentachlorophenol : Chemistry, Pharmacology and Environmental Toxicology, (ed) Rao K R, Environmental Science Research No 12, 1978, Plenum Press, New York

Pignatello J J, Martinson M M, Steiert J G, Carlson R E and Crawford R L (1983) **Biodegradation and Photolysis of Pentachlorophenol in Artificial Freshwater Streams,** Appl Environ Microbiol 46 (5), 1024 - 1031

Pionke H B and Glotfelty D E (1989) **Nature and Extent of Groundwater Contamination by Pesticides in an Agricultural Watershed,** Wat Res Vol 23, No 8, 1031 - 1037

Portmann JE (1975) **the Bioaccumulation and Effects of Organochlorine Pesticides in Marine Animals,** Proc R Soc Lond B 189, 291-304

Portmann JE (1979) **the Bioaccumulation and Effects of Organochlorine Pesticides in Marine Animals,** Proc R Soc Lond B 189, 291-304

Prasad T A V, Srinivas T, Rafi F Md and Reddy R C (1991) **Effect in Vivo of Atrazine on Haematology and O_2 Consurption in Fish,** *Tilapia mossambica,* Biochem Internat 23, (1), 157 - 161

Pyysalo H, Wickström K, Litmanen R, Lindström-Seppä P, Koivusaari U and Hänninen O (1981) **Contents of Chlordane, PCB and DDT Compounds and the Biotransformation Capacity of Fishes in the Lake Area of Eastern Finland,** Chemosphere 10, 865-876

Rajendran N and Venugopalan UK (1991) **Bioconcentration of Endosulfan in Different Body Tissues of Estuarine Organisms under Sublethal Exposure,** Bull Environ Contam Toxicol 46 (1), 151 - 158

Rao PSC and Davidson JM (1980) **Estimation of Pesticide Retention and Transformation Paramters Required in Non-point Source Pollution Models,** In: Overcash M and Davidson J (Eds) Environmental Impact of Non-point Source Pollution, Ann Arbor Science Publishers, Ann Arbor, MI pp 23-68

Rand GM and Petrocelli SR (1985) **Fundamentals of Aquatic Toxicity,** Hemisphere Publishing Corporation, New York pp 542

Rawn G P, Webster G R B and Muir D C G (1982) **Fate of Permethrin in Model Outdoor Ponds,** J Environ Sci Health B17, 463 - 486

Reed RJ (1966) **Some Effects of DDT on the Ecology of Salmon Streams in Southeastern Alaska,** US Fish Wildl Serv Spec Scient Rep, Fisheries, No 542, pp 15

Reinert RE (1967) **The Accumulation of Dieldrin in an Alga (*Scenedesmus obliquus*) daphnia (*Daphnia magna*), guppy (*Lebistes reticulatus*) food chain,** PhD thesis, University of Michigan, Ann Arbor, Michigan pp76

Reinert RE (1969) **Insecticides and the Great Lakes,** Limmos Magazine (US), 2(3), 4-9

Reinert RE (1970) **Pesticide Concentrations in Great Lakes Fish,** Pestic Monit J, 3, 233-40

Reinert R E (1972) **Accumulation of Dieldrin in an Algae (*Scenedesmus obliquus*), *Daphnia magna* and the Guppy (*Poecilia reticulata*),** J Fish Res Bd Canada 29, 1413 - 1418

Reinert RE, Stone LJ and Willford WA (1974) **Effect of Temperature on Accumulation of Methylmercuric Chloride and p,p'DDT by Rainbow Trout (*Salmo gairdneri*),** J Fish Res Bd Canada, 31, 1649-52

Reinert RE and Bergman HL (1974) **Residues of DDT in Lake Trout (*Salvelinus namaycush*) and Coho Salmon (*Oncorhynchus kisutch*) from the Great Lakes,** J Fish Res Can 31, 191-199

× Reish D J and Lemay J A (1991) **Toxicity and Bioconcentration of Metals and Organic Compounds by Polychaeta,** Ophelia Suppl 5, 653 - 660

Reynoldson TB and Davies RW (1980) **A Comparative Study of Weight Regulation in *Nephelopsis obscura* and *Erpobdella punctata* (Hirudinoidea)** Comp Biochem Physiol 66A, 711-714

Rice C P and Sikka H C (1973) **Fate of Dieldrin in Selected Species of Marine Algae,** Bull Environ Contam Toxicol Vol 9, 116 - 123

Rickard D G and Dulley M E R (1983) **The Levels of Some Heavy Metals and Chlorinated Hydrocarbons in Fish From the Tidal Thames,** Environ Pollut Ser B 5, 101 - 119

Risebrough R W, Menzel D B, Martin D T and Olcott H S (1967) **DDT Residues in Pacific Sea Birds: a Persistent Insecticide in Marine Food Chains,** Nature, Lond 216, 589 - 590

Roberts D (1975c) **The Effect of pesticides on Byssus Formation in the Common Mussel, *Mytilus edulis*,** Environ Pollut 8, 241-254

Robinson J, Richardson A, Crabtree A N, Coulson J C and Potts G R (1967) **Organochlorine Residues in Marine Organisms,** Nature Vol 214, 1307 - ???

Romeril MG (1979) **The Occurrence of Copper, Iron and Zinc in the Hard Shell Clam, *Mercenaria mercenaria* and Sediments of Southampton Water,** Estuar Coastl Mar Sci 9, 423-434

Rosenberg OM (1975) **Food Chain Concentration of Chlorinated Hydrocarbon Pesticides in Invertebrate Communities: A Re-evaluation,** Quaest Ent 11, 97-100

Rubinstein NI, Lores E and Gregory NR (1983) **Aquat Toxicol 3, 249**

Ryan JJ, Lau PY, Pilon JC, Lewis D, McLeod HA and Gervals A (1984) **Incidence and Levels of 2,3,7,8 -tetrachlorodibenzo-p-dioxin in Lake Ontario Commercial Fish,** Environ Sci Technol 18, 719-721

Salazar S M et al (1987) **Effects of TBT on Marine Organisms: Field Assessment of a New Site-specific Bioassay System.** In: Proceedings of Oceans 87 (Marine Technology Society and IEEE Ocean Engineering Society) pp 1461 - 1470. Halifax Nova Scotia, Canada,

Sanders H O and Chandler J H (1972) **Biological Magnification of a Polychlorinated Biphenyl (Aroclor 1254) from Water by Aquatic Invertebrates,** Bull Environ Contam Toxicol 7, 257 - 263

✗ Say P J, Harding J P C and Whitton B A (1981) **Aquatic Mosses as Monitors of Heavy Metal Contamination in the River Etheraw, Great Britain,** Environ Pollut Ser B 2, 295 - 307

✗ Say P J & Whitton B A (1983) **Accumulation of Heavy Metals by Aquatic Mosses 1: *Fontinalis antipyretica* Hedw,** Hydrobiological 100, 245 - 260

✓ Say P J, Burrows I G and Whitton B A (1990) ***Enteromorpha* as a Monitor of Heavy Metals in Estuaries,** Hydrobiologia 195, 119-126

Schafer H A, Hershelman G P, Young D R and Mearns A (1982) **Contaminants in Ocean Foodwebs,** In: Southern California Coastal Research Project Biennial Report 1981-82 (W Basco Ed) 17-28 Long Beach, California

Schnare D W, Ben M and Shields M G (1984) **Body Burden Reductions of PCBs, PBBs and Chlorinated Pesticides in Human Subjects,** Ambio Vol 13 No 34

Schuurmann G and Klein W (1988) **Advances in Bioconcentration Prediction,** Chemosphere 17 (8), 1551 - 1574

Sharom M S and Solomon K R (1981) **Adsorption-desorption, degradation and distribution of permethrin in aqueous systems,** J Agric Food Chem 29, 1122 - 1125

Shaw G R and Connell D W (1982) **Factors Influencing Concentrations of Polychlorinated Biphenyls in Organisms from an Estuarine Ecosystem,** Aust J Mar Freshwater Res 33, 1057 - 1070

Shaw G R and Connell D W (1984) **Physiochemical Properties Controlling Polychlorinated Biphenyl (PCB) Concentrations in Aquatic Organisms,** Environ Sci Technol 18, 18 - 23

Shaw GR and Connell DW (1987) **Comparative Kinetics for Bioaccumulation of Polychlorinated Biphenyls by the Polychaete (*Capitella capitata*) and Fish (Mugil cephalus),** Ecotoxic Envir Saf 13, 84-91

Sheridan PF (1975) **Uptake, Metabolism and Distribution of DDT in Organs of the Blue Crab, *Callinectes sapidus*,** Chesapeake Sci 16, 20-6

Short J W and Thrower F P (1986) **Accumulation of Butyltin in Muscle Tissue of Chinook Salmon Reared in Sea Pens Treated with Tri-n-butyltin.** In: Proceedings of Ocean 86 (Marine Technology Society and IEEE Ocean Engineering Society) pp 1177 - 1181, Washington DC

Smith BS (1971) **Sexuality in the American Mud-snail, *Nassarius Obsoletus* Say,** Proceedings of the Malacological Society of London 39, 377-378

Smith BS (1981c) **Male Characteristics in Female *Nassarius obsoletus*: Variations Related to Locality, Season and Year,** Veliger 23, 212-216

Smith P and Smith L (1975) **Organotin Compounds and Applications,** Chemistry in Britain 11, 208 - 12 and 226

Södergren A (1968) **Uptake and Accumulation of C^{14} - DDT by Chlorella sp (Chlorophyceae)** Oikos 19, 126

Södergren A, Svensson B and Ulfstrand S (1972) **DDT and PCB in South Swedish Streams,** Environ Pollut 3, 25 - 36

Södergren A and Svensson B (1973) **Uptake and Accumulation of DDT and PCB by *Ephmara danica* (Ephemeroptera) in Continuous - Flow Systems,** Bull Environ Contam Toxicol 7, 257 - 263

Södergren A and Larsson P (1982) **Transport of PCBs in Aquatic Laboratory Model Ecosystems form Sediment to the Atmosphere via the Surface Microlayer,** Ambio, 11, 41-5

Spague JB, Elson PF and Duffey JR (1971) **Decrease in DDT Residues in Young Salmon after Forest Spraying in New Brunswick,** Environ Pollut 1, 191-202

Srinivas T, Prasad T A V, Raffi G Md and Reddy D C (1991) **Effect of Atrazine on Some Aspects of Lipid Metabolism in Fresh Water Fish,** Biochem Internat 23 (3), 603 - 609

Staples CA, Dickson KK, Rodgers JH Jr and Saleh TY (1983), **A Model for Predicting the Influence of Suspended Solids on the Bioavailability of Neutral Organic Chemicals in the Water Compartment,** Presented at the Seventh Annual ASTM Symposium on Aquatic Toxicology Milwaukee, Wisconsin, 18-19 April 1983

Stegeman JJ, Klotz AV, Woodin BR and Pajor AM (1981) **Induction of Hepatic Cytochrome P_{450} in Fish and the Indication of Environmental Induction in Scup,** Aquat Toxicol 1, 197-212

Stein J H, Hom T and Varanasi U (1984) **Simultaneous Exposure of English Sole** *(Parophrys vetulus)* **to Sediment-associated Xenobiotics: Part 1. Uptake and Disposition of 14 C-polychlorinated Biphenyls and 3 H-benzo (a) Pyrene,** Mar Environ Res 13, 97 - 119

Streit B (1979) **Uptake, Accumulation and Release of Organic Pesticides by Benthic Invertebrates, 2 Reversible Accumulation of Lindane, Paraquat and 2, 4 -D from Aqueous Solution by Invertebrates and Detritrus,** Arch Hydrobiol Suppl 55, 349-372

Streit B and Schwoerbel J (1976) **Experimentelle Untersuchungen über die Akkumulation und Wirkung von Herbiziden bei Benthischen Süsswassrtieren,** Verh Ges f Okologie Göttingen 371-383

Streit B and Peter H-M (1978) **Long Term Effects of Atrazine to Selected Freshwater Invertebrates,** Arch Hydrobiol Suppl 55, 62 - 77

Strek H J and Weber J B (1982) **Behaviour of Polychlorinated Biphenyls (PCBs) in Soils and Plants,** Environ Pollut Ser A 28, 291-312

Swartz RC and Lee H II (1980) in RA Baker (Ed), **Contaminants and Sediments, Vol 2, Analysis, Chemistry, Biology,** Ann Arbor Science Publishers Inc, The Butterworth Group, Ann Arbor, MI, 533-553

Tanabe S, Tatsukawa R and Phillips D J H (1987) **Mussels as Bioindicators of PCB Pollution: A Case Study on Uptake and Release of PCB Isomers and Congeners in Green-lipped Mussels** *(Perna viridis)* **in Hong Kong Waters,** Environ Pollut 47, 41 - 62

Tarzwell C M (c1965) **The Toxicity of Synthetic Pesticides to Aquatic Organisms and Suggestions for Meeting the Problem**

Ten Berge WF and Hillebrand M (1974) **Organochlorine Compounds in Several Marine Organisms from the North Sea and the Dutch Wadden Sea,** Neth J Sea Res 8, 361-8

Tenore KR, Goldman JC and Clarner JP (1973) **The Food Chain Dynamics of the Oyster, Clam and Mussel in an Aqua Culture Food Chain,** J Exp Mar Biol and Ecol 12, 157-167

Tesch F-W (1977) **The Eel, Biology and Management of Anguillid Eels,** Chapman and Hall Ltd, Edinburgh, pp 434

Thyband E and Caquet T (1991) **Uptake and Elimination of Lindane by** *Lymnaea palustris* **(Mollusca: Gastropoda) : A Pharmacokinetic Approach,** Ecotox Environ Safety 21, 365 - 376

Tobby T E and Durbin F J (1975) **Lindane Residue, Accumulation and Elimination in Rainbow Trout and Roach.** Environ Pollut 8, 79 - 89

Unsal M (1984) **Accumulation and Loss of Tin by the Mussel,** Oceanol Acta 7, 493 - 498

US Dept of Interior: Fish and Wildlife Service, **Monitoring Fish and Wildlife for Environmental Contaminants,** The National Contaminant Biomonitoring Program

US Dept of Interior: Fish and Wildlife Service, **National Pesticide Monitoring Program: Organochlorine Residues in Freshwater Fish, 1979 - 1979,** Resource Publication 152

Valkins AO, Seligman PF and Lee RF (1986) **Butyltin Partitioning in Marine Waters and Sediment,** In: Oceans '86 Proceedings of Organotin Symposium IEEE Inc Washington DC

van den Broek W L F (1979) **Seasonal Levels of Chlorinated Hydrocarbons and Heavy Metals in Fish and Brown Shrimps from the Medway Estuary, Kent,** Environ Pollut 19, 21 - 37

van der Kerk, G J M (1970) **Present Trends and Perspectives in Organotin Chemistry,** Chemistry and Industry 644 - 647

van der Oost R, Heida H and Opperhuizen A (1988) **Polychlorinated Biphenyl Congeners in Sediments, Plankton, Mollusks, Crustaceans and Eel in a Freshwater Lake - Implications of Using Reference Chemicals and Indicator Organisms in Bioaccumulation Studies,** Arch Environ Contam Toxicol 17 (6), 721 - 729

van Zoest R and van Eck G T M (1990) **Behaviour of Particulate Polychlorinated Biphenyls and Polycyclic Aromatic Hydrocarbons in the Scheldt Estuary,** Neth J Sea Res 26 (1), 89 - 96

van Zoest R, van Eck G T M (1991) **Occurrence and Behaviour of Several Groups of Organic Micropollutants in the Scheldt Estuary,** Sci Tot Environ 103, 57 - 71

Veith GD, DeFoe DFL and Bergstedt BV (1979) **Measuring and Estimating the Bioconcentration Factor in Fish,** J Fish Res Board Can 36, 1040-1048

Vreeland V (1974) **Uptake of Polychlorinated Biphenyls by Oysters,** Environ Pollut 6, 135 - 140

Waddington JI and MacKay DW (1972) **Pollution Control in Estuaries and Inshore Waters,** Inst Wat Pollut Contol Symp on River Pollution Prevention, p17

Waldock M J, Thain J E and Miller D (1983) **The Accumulation and Depuration of bis (Tributyltin) Oxide in Oysters: A Comparison between the Pacific Oyster (*Crassostrea gigas*) and the European Flat Oyster (*Ostrea edulis*)** ICES, CM 1983 / E : 52

Waldock MJ and Miller D (1983) **The Determination of Total and Tributyltin in Sea Water and Oysters in Areas of High Pleasure Craft Activity,** ICES Paper CM 1983/E:12 Int Council Explor Sea, Copenhagen

Waldock M J, Thain J E and Waite M E (1987) **The Distribution and Potential Toxic Effects of TBT in UK Estuaries during 1986,** Appl Organomet Chem, 1, 287 - 301

Waldock M J, Waite ME and Thain JE (1988) **Inputs of TBT to the Marine Environment from Shipping Activity in the UK,** Environ Technol Letters 9, 999 - 1010

Walker C H (c1975) **The Persistence and Toxicity of Pesticides in Relation to Enzyme Activity,** from: Outlook in Agriculture

Walker C H (1987) **Kinetic Models for Predicting Bioaccumulation of Pollutants in Ecosystems,** Environ Pollut 44, 227 - 240

Wang T (1991) **Assimilation of Malathion in the Indian River Estuary, Florida,** Bull Environ Contam Toxicol 47 (2) 238 - 243

Ware GW, Dee MK and Cahill WP (1968) **Water Flora as Indicators of Irrigation, Water Contamination by DDT,** Bull Environ Contam Toxicol 3, 333-338

Water Research Centre (1988) **Information Related to the Proposed "Red List" substances,** PRU 1901- M/2

Wathern P, Young S N, Brown I W and Roberts D A (1987) **UK** Interpretation and Implementaion of the EEC Shellfish Directive

Webster EJ (1967) **An Autoradiographic Study of Invertebrate Uptake of DDT-Cl^{36},** Ohio J Sci, 67, 300-307

Wegman R C C and van den Broek H H (1983) **Chlorophenols in River Sediment in The Netherlands,** Wat Res 17, 227 - 230

Wehr J D and Whitton B A (1983) **Accumulation of Heavy Metals by Aquatic Mosses, 2: *Rhynchostegium riparioides*,** Hydrobiologia 100, 261-284

Welsh Water Authority (Jan 1987) Polychlorinated Biphenyls (PCBs) in Fish from Caerphilly Castle Moat

Wharf JR (1975) **Ecological Studies of the Benthic Invertebrate Macrofauna of the Lower Medway Estuary (Kent), with Special Reference to the Occurrene of Specific Pollutants**, PhD thesis, University of London

Wharfe J R, van den Broek W L F (1978) **Chlorinated Hydrocarbons in Macroinvertebrates and Fish from the Lower Medway Estuary, Kent** Mar Pollut Bull 9, 76 - 79

Wheeler W B (1970) J Agri Food Chem 18, 46

Whitton B A (1991) **Aims of Monitoring**, In: Whitton B A, Rott E and Friedrich G (Eds) Use of Algae for Monitoring Rivers, Proceedings of an International Symposium, Düsseldorf, Germany 26-28 May 1991, Rott E Publisher, Innsbruck

Whitton BA, Burrows IG and Kelly MG (1989) **Use of *Cladophora glomerata* to Monitor Heavy Metals in Rivers**, J Appl Phycol I, 293-299

Whitton BA, Kelly MG, Harding JPC and Say PJ (1991) **Use of Plants to Monitor Heavy Metals in Freshwaters 1991**, Standing Committee of Analysts, HMSO, London

Wierinck I and Verstraete W (1990) **Degradation of Atrazine by a Hydrogenotrophic Microbial Association**, Environ Technol 11, 843 - 852

Widdows J and Donkin P (1989) **The Application of Combined Tissue Residue Chemistry and Physiological Measurements of Mussels (*Mytilus edulis*) for the Assessment of Environmental Pollution**, Hydrobiologia 188/189, 455-461

Wildish DJ and Zitko V (1971) **Uptake of Polychlorinated Biphenyls from Sea Water by *Gammarus oceanicus*,** Mar Biol 9, 213-218

Woodwell GM (1967) **Toxic Substances and Ecological Cycles**, Scientific American 216 (3) 24-31

Woodwell GM, Wurster CF and Isaacson PA (1967) **DDT Residues in an East Coast Estuary: A Case of Biological Concentration of a Persistent Insecticide**, Science NY 156, 821-824

Young DR, Heesen TC and McDermott DJ (1976) **An Offshore Biomonitoring System for Chlorinated Hydrocarbons**, Mar Pollut Bull 7, 156-159

Young D R, McDermott-Ehrlich D and Heesen T C (1977) **Sediments as Sources of DDT and PCB**, Mar Pollut Bull 8, 245 - 257

Young DR, Mearns AJ, Jan TK, Heesen TC, Moore MD, Eganhouse RP, Hershelman GP and Gosset RW (1980), **Trophic Structure and Pollutant Concentrations in Marine Ecosystems of Southern California**, California Cooperative Oceanic Fisheries Investigation Report, 21, 197-206

Zaroogian GE (1980) **Crassostrea Virginica as an Indicator of Cadmium Pollution**, Mar Biol 58, 275-284

Zierath DL, Hassett JJ and Banwart WL (1980), Soil Sci, 129, 277

Zitko V (1971) **Polychlorinated Biphenyls and Organochlorine Pesticides in Some Freshwater and Marine Fishes,** Bull Environ Contam Toxicol, 6, 464-70

Zuolian C and Jensen A (1989) **Accumulation of Organic and Inorganic Tin in Blue Mussel,** ***Mytilus edulis*** **Under Natural Conditions,** Mar Pollut Bull 20, 281 - 286