



### Working Papers Prepared as Background for Testing for Effects of Chemicals on Ecosystems (1981)

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WORKING PAPERS  
PREPARED AS BACKGROUND FOR  
TESTING FOR EFFECTS OF CHEMICALS ON ECOSYSTEMS

Prepared for the  
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## PREFACE

The eleven working papers in this volume were prepared for the study of the Committee to Review Methods for Ecotoxicology. The charge to the Committee was to identify characteristics of ecological systems that would indicate hazardous effects of chemicals beyond the level of single species, to establish criteria for suitable testing schemes, and to evaluate the effectiveness of available test systems in assessing effects of chemicals within ecosystems. To assist in its deliberations on the broad range of issues to be addressed, the Committee sought additional input from a number of experts. The working papers address a variety of topics: the use of microcosms as a testing scheme in terrestrial and aquatic systems; lethal gene distribution and diatoms as monitoring techniques for hazard assessment of chemicals in ecosystems; special problems associated with hazard assessment in wetlands and watersheds; approaches to assessing the environmental impact of radionuclide and xenobiotic organic substances; a general review of ecosystem properties relevant to ecotoxicology; and a discussion of the advantages and disadvantages of various classes of ecological tests.

The Committee benefited greatly from these contributions and wishes to thank each of the authors.

John Cairns, Jr.  
Chairman  
Committee to Review Methods  
for Ecotoxicology

## CONTENTS

<p>✓ TERRESTRIAL MICROCOSMS - A STATE OF THE ART REVIEW                ✓ James Berg</p>	<p>1</p>
<p>    Introduction</p>	<p>2</p>
<p>    Processes and Properties Most Transferrable to Real World Situations</p>	<p>2</p>
<p>    Uses of Microcosms in Ecological Research</p>	<p>3</p>
<p>    Life Span of Microcosms</p>	<p>6</p>
<p>    Limitations</p>	<p>6</p>
<p>    Literature</p>	<p>9</p>
<p>✓ LAKES AND MICROCOSMS: EXTENDING MICROCOSM DATA TO AQUATIC ECOSYSTEMS                ✓ Peter H. Gleick</p>	<p>17</p>
<p>    Abstract</p>	<p>17</p>
<p>    Introduction</p>	<p>18</p>
<p>    Microcosm Dynamics</p>	<p>20</p>
<p>    Micro-Variable Effects on Macro-Variables</p>	<p>22</p>
<p>    Complicating Factors</p>	<p>34</p>
<p>    Summary</p>	<p>42</p>
<p>    References</p>	<p>44</p>
<p>✓ OPTIMUM MICROCOSMS FOR LAKE ECOTOXICOLOGY                ✓ John Harte</p>	<p>51</p>
<p>    Introduction</p>	<p>52</p>
<p>    Design</p>	<p>52</p>
<p>    Initiation</p>	<p>54</p>
<p>    Operation</p>	<p>60</p>
<p>    Conclusion</p>	<p>66</p>
<p>    References</p>	<p>68</p>
<p>✓ ON THE FEASIBILITY OF USING LETHAL + SEMILETHAL FREQUENCIES AND THE DISTRIBUTION OF LETHALS IN <u>DROSOPHILA MELANOGASTER</u> NATURAL POPULATIONS AS A MONITORING TECHNIQUE FOR THE DETECTION OF CHEMICAL MUTAGENS IN THE ENVIRONMENT                ✓ P.T. Ives and H.T. Band</p>	<p>71</p>
<p>    Abstract</p>	<p>72</p>
<p>    X-chromosome Lethals and the Problem Posed by Hi Mutator Genes in Populations</p>	<p>74</p>
<p>    Second Chromosome Lethals and the Dual Problems Posed by Changes in Both Frequencies and Distribution Over Time</p>	<p>75</p>
<p>    The Populations Sampled</p>	<p>76</p>
<p>    The Weather Data Available in the Amherst Area</p>	<p>77</p>
<p>    The South Amherst Population in the 1970's</p>	<p>78</p>
<p>    Literature</p>	<p>83</p>
<p>    Tables and Figures</p>	<p>87</p>

✓	CHEMICALS AND WETLANDS	97
	✓ R.H. Kadlec	
	Abstract	97
	Background	98
	Assessment Goals	99
	Wetland Features Relevant to Chemical Interactions	102
	Heavy Metals	105
	Refractory Chemicals	107
	Other Water Quality Parameters	108
	Pathogens	110
	Conclusions	112
	References	114
✓	ECOSYSTEM PROPERTIES RELEVANT TO ECOTOXICOLOGY	119
	✓ Richard Levins	
	Connectivity	119
	Random Processes	121
	Nonlinear Responses	122
	Natural Selection	125
	Tables and Figures	127
✓	THE UTILITY OF DIATOMS FOR HAZARD ASSESSMENT OF CHEMICALS IN ECOSYSTEMS	133
	✓ Rex L. Lowe	
✓	ECOTOXICOLOGY AT THE WATERSHED LEVEL	137
	✓ Logan A. Norris	
	Direct and Indirect Effects	138
	Perspectives on Perturbations	140
	Information Needed to Assess Effects of Chemicals at the Watershed Level of Resolution	141
	Factors Affecting the Behavior of Chemicals in Watersheds	143
	Chemicals of Concern	144
	Features of Watersheds Which Influence Toxicity	146
	References	147
✓	THE UTILITY OF SINGLE SPECIES AND ECOSYSTEM TESTS IN ASSESSING THE ENVIRONMENTAL IMPACT OF RADIONUCLIDE ECOTOXICANTS	149
	✓ Harvey L. Ragsdale	
	Introduction	150
	Modes of Entry	151
	Points of Impact	153
	Temporality	162
	Discussion	163
	Literature	166

✓ CLASSES OF ECOTOXICOLOGICAL TESTS: THEIR ADVANTAGES AND DISADVANTAGES FOR REGULATION Richard A. Schneider	175
Abstract	175
Introduction	176
Single-Species Tests	177
Microcosm Tests	180
Field Tests	183
Evaluation of the Test Types, Summary, and Conclusions	185
References	189
✓ AN ECOSYSTEM APPROACH TO THE TOXICOLOGY OF RESIDUE FORMING XENOBIOTIC ORGANIC SUBSTANCES IN THE GREAT LAKES Wayland R. Swain	193
Figure 1: A Conceptual Framework for Toxic Substances in the Great Lakes Ecosystem	196
Source Inputs of Xenobiotic Compounds	197
Table 1: PCB's in Precipitation in the Great Lakes Basin	198
Table 2: Mean Concentrations of Toxic Substances and Pesticides in Rain and Snowfall	200
Table 3: Mass Balance of PCB in Lake Superior Accumulations in Great Lakes Water Masses, Plankton, and Sediment	202
Table 4: Polychlorinated Biphenyl Compounds Observed in Waters of the Great Lakes	204
Table 5: Organochlorine Substances Observed in the Waters of the Great Lakes	206
Table 6: Occurrence of Chlorinated Hydrocarbons in the Plankton, <u>Cladophora</u> , and Benthic Fauna of Lake Ontario	211
Table 7: Concentrations ( $\mu\text{g/g}$ dry weight) of Chlorinated Hydrocarbons in Lake Ontario Net Plankton ( $64 \mu$ mesh) - 1975 Accumulation in Great Lakes Fish	213
Table 8: Mass Sedimentation Rates in the Great Lakes	214
Table 9: Organochlorine Compounds in the Sediments of the Great Lakes	215
Adult Human Exposure	220
Table 10: Polychlorinated Biphenyls Observed in Fish of the Great Lakes	221
Table 11: Representative Mean Contaminant Ranges in Selected Great Lakes Fish	229
Table 12: Calculated PCB Exposure Potential for Non-Urban Great Lakes Basin Residents from Drinking Water and Respiration Effects of Acute Exposure	232
Figure 2: Frequency Distribution of Reported Great Lakes Fish Consumption, Exposed Population	233
Effects of Chronic Exposure	234
Accumulation in Adipose Tissue	234
Accumulation in Blood	235
Intermediate Effects of Consumption of Great Lakes Fish	235
Table 13: PCB Residues (mg/kg) in Human Adipose Tissue	236

**Terrestrial Microcosms - A State of the Art Review**

**A Report to the National Academy of Sciences - Committee on Ecotoxicology**

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Graduate Student  
The University of Tennessee  
Graduate Program in Ecology**

**February 18, 1980**

## I. Introduction

Terrestrial microcosms, or microecosystems, have long been used as attempts to simplify studies of natural ecosystems (Odum, 1971). Traditionally, this has been done by placing biota and appropriate substrata under controlled environmental conditions in closed containers. By controlling many physical parameters, the effects of selected variables upon the biota can be assessed. The fundamental objective is experimental manipulation of entire ecosystems, or representative subsets.

As new techniques and instruments have been developed, new applications of microcosms have arisen; and the use of microcosms as research tools has proliferated (Anonymous, 1976; Ausmus, et al, 1977; Bondietti, et al, 1976; Draggan, 1976 a, 1976 b; Draggan & Giddings, 1978; Draggan & Van Voris, 1979; Gillett & Witt, 1977; McCormick, et al, 1974; Sanders, 1976; Witt & Gillett, 1977).

The complex physical structure of terrestrial ecosystems presents significant problems seldom encountered in aquatic ecosystems. Aquatic ecosystems have the advantage of being composed primarily of species which are small in size, which occur in high densities, and which have rapid turnover rates. These characteristics are desirable when attempting to develop microcosms, and they are seldom found in terrestrial ecosystems (Platt & McCormick, 1964).

## II. Processes and Properties Most Transferrable to Real World Situations

Microcosms share with one another, and with their real world counterparts, basic ecological processes and properties. These have been identified by the NAS-NRC Committee on Ecotoxicology. It is difficult or impossible to compare the behavior of the variable species which comprise microcosms, but it is relatively easy to compare the processes which most microcosms and natural systems share.

Results of recent workshops conducted by ORNL, at the request of EPA, substantiate the conclusions of earlier workers (Van Voris, et al, in press) who identified fluxes of nutrients and CO<sub>2</sub> as the most useful parameters for monitoring the behavior of terrestrial microcosms. Additional parameters of value in specific situations include productivity, diversity, enzyme activity, ATP content, chlorophyll ratios, and dissolved organic carbon (Ausmus & O'Neill, 1977; Coleman, et al, 1978 a; Elliott, et al, 1979; Van Voris, et al, in press). The preferred method of monitoring CO<sub>2</sub> flux is by infra-red gas analysis (Ausmus & O'Neill, 1977; Ausmus, et al, 1977; Murphy, 1970; Murphy & McCormick, 1971; Olson, 1975; Lugo, 1969). Methods of measuring nutrient flux must vary to accommodate variable response time of different systems (Ausmus, et al, 1977; Ausmus & O'Neill, 1977; Coleman, et al, 1978 a; Van Voris, et al, in press).

### III. Uses of Microcosms in Ecological Research

A. Microcosms are most generally used to either: a) validate hypotheses or models of ecosystem processes and behavior, or b) to identify cause and effect relationships between environmental influences and ecosystem properties, or c) to identify the fate and effects of toxins or other elements in ecological systems. An example of the former is the use of microcosms to test hypotheses of ecosystem stability which cannot be studied in larger natural ecosystems (Van Voris, et al, in press; Patten & Witkamp, (1967). Examples of the second type of use include the work of Gile & Gillett (1979) and Nabholz (1978), on the ecological effects of toxins; the work of Murphy & McCormick (1971) on the effects of beta radiation from simulated radioactive fallout, and the studies of Olson (1975) on acid rain. The use of microcosms to examine the fate of toxins is especially widespread (Gillett & Gile, 1976; Metcalf & Lu, 1973; Lichtenstein, 1977; Gillett, et al, 1974).

B. Gnotobiotic microcosms are perhaps the least complex of microcosms, but they are usually the most difficult to maintain. By carefully selecting combinations of species, substrata and environment we lose the benefit of long term trial and error provided by natural selection. Even in nearly closed systems as simple as a few microbial species, glass microbeads, and a solution of mineral salts (Anderson & Coleman, 1977; Draggan, 1976 c), population interactions and system homeostasis may be greatly affected by the sequence of introduction of components. Initial low costs of developing gnotobiotic systems may soon be overcome by the cost of increasingly sophisticated technology required to maintain gnotobiotic systems with increasing trophic levels.

Gnotobiotic systems have been especially valuable in gaining insight to predator-prey relationships, nutrient cycling and the co-existence of apparently competing species (Anderson, et al, 1977; Anderson, et al, in press; Anderson, et al, 1978; Anderson, et al, 1979; Cole, et al, 1978; Coleman, et al, 1977; Coleman, et al, 1978; Coleman, et al, 1978; Herrera, et al, 1978; Herzberg, et al, 1978). While the advantage of gnotobiotic microcosms is the opportunity to examine specific interactions, a major deficiency is the lack of real world complexity. Gnotobiotic microcosms permit greater precision of measurement but the process and properties being measured may be unrealistically simplified.

C. Fragments of terrestrial ecosystems are often employed in a manner similar to using sub-samples of aquatic ecosystems. These microcosms typically consist of soil cores or homogenized soil and litter in a relatively closed container. Gaseous inputs and outputs are monitored to estimate metabolic activity and leachates are monitored for determining mineral nutrient fluxes and dissolved organic carbon. Typically, once baseline data are obtained, the "black box" is placed in a new environment or is disrupted to simulate a desired impact (Ausmus & O'Neill, 1977; Bond, et al, 1976; Bond, et al, 1977; Jackson, et al, 1977; Jackson & Hall, 1978; Klein,

et al, 1972; Kudeyarov & Jenkinson, 1976; Lichtenstein, et al, 1977; Lighthart, et al, 1977; Lighthart & Bond, 1976; Odum & Lugo, 1970; Snyder & Wullenstein, 1973; Spalding, 1978; Tu, 1978; Vissner, et al, 1973; Witkamp & Frank, 1970; Zentmeyer, 1955).

These systems have been especially valuable in monitoring the fate, pathways, residence times, and metabolic products of toxic chemicals (Atlas, et al, 1978; Ausmus, et al, 1979; Bartha & Pramer, 1965; Coats, et al, 1976; Gordon, et al, 1969; Kaufman, 1977; Klein, 1977; O'Neill et al, 1977; Walter - Echols & Lichtenstein, 1977 & 1978; Witkamp & Frank, 1969).

D. Whole microecosystems seldom possess the properties desired of microcosms. However, when natural terrestrial ecosystems are identified which have these properties (McCormick, et al, 1974), they offer two significant advantages: 1) even when experimentally manipulated, they more closely mimic real world phenomena, and 2) they can be used either as open or closed systems depending upon experimental objectives. One way of demonstrating transferability of microcosm behavior to real world field situations is to compare microecosystems under field conditions with whole system microcosms under controlled conditions, and/or with fragments of these systems under rigidly controlled conditions (Lugo 1969; Meyer, et al, 1975; Murphy & McCormick, 1971; Shartz & McCormick, 1973). Whole system microcosms have been used to test for the effect of a variety of pesticides, herbicides, fungicides, and biocides (Anderegg, et al, 1977; Beall, et al, 1976; Cole & Metcalf, 1977; Cole, et al, 1976; Fuhremann & Lichtenstein, 1978; Gile & Gillett, 1979 a&b; Gillett & Gile, 1976; Hirwe, et al, 1975; Kapoor, et al, 1970; Kapoor, et al, 1972; Klein, et al, 1975; Kuntsman & Lichtenstein, 1979; Lichtenstein, 1977; Lichtenstein, et al, 1967; Lichtenstein, et al, 1978; Lichtenstein, et al, 1977; Lu, et al, 1975; Metcalf, et al, 1971; Sanborn, et al, 1976).

Microcosms provide the containment essential to the safe use of radionuclides in tracking ecological pathways of elements. The combined use of microcosms and radiolabeled chemicals to delineate time courses, ecological pathways, and effects of toxic substances offers one of the most promising areas of ecotoxicology research (Ausmus, et al, 1977; Ausmus, et al, 1978; Draggen, 1976 a&c; Jackson, et al, 1978 a&b; Jackson, et al, 1979; Jackson & Levin, 1979; Patten & Witkamp, 1967; Van Voris, et al, in press; Witkamp, 1976; Witkamp & Ausmus, 1975).

#### IV. Life Span of Microcosms

There is an initial period of several days to a few weeks during which newly synthesized or recently transplanted systems become acclimated to the experimental environment. A second period of approximately the same duration is required to accumulate baseline data for the properties being monitored. It is especially important to identify rhythms and cycles during this period (Van Voris, Battelle, Columbus). Characteristically, the third stage is one of experimental manipulation. Longevity of microcosms appears to be related to the response times of ecosystem processes. Application of toxins or other stresses must be timed according to the periodicity of system activities. Measurements of responses must be carefully scheduled to accommodate response times of the processes being measured. If manipulations are non-destructive, experimental and control microcosms may persist for years. Using the techniques of Platt & McCormick (1964), whole microecosystems have been maintained for study for as long as ten years.

#### V. Limitation

The best materials and techniques for conducting microcosm research are expensive. It is expensive to maintain large areas under rigidly controlled environmental conditions. Glass containers of prescribed composition are also expensive. Once problems of container surface interactions or light

transmission are resolved, one must still contend with time consuming and expensive means of monitoring gas and liquid inputs and outputs (J. D. Gile, EPA, CERL, Corvallis, OR).

The limited size of terrestrial microcosms creates problems of reproducibility, intra and inter-system variance and constrained mobility of organisms. As microcosm size is increased to accommodate these constraints, cost increases significantly.

A significant shortcoming of closed microcosms is the loss of continuous inputs of propagules, inorganic, and organic matter which characterize natural open terrestrial ecosystems. The larger and more open the microcosm, the longer they persist and the more closely they mimic natural processes. There is some evidence to the contrary for aquatic microcosms (Giddings & Eddleman, 1977). Smaller microcosms usually allow for greater number of replications, more uniform environmental conditions, and more precise monitoring.

Increased activity in theoretical and mathematical ecology is responsible for much of the growing use of microcosms (Patten & Witkamp, 1967). Terrestrial microcosms can serve as fast response systems for testing equations or models which attempt to explain specific interactions. Terrestrial microcosms are most useful in describing process characteristics of whole systems. As we learn more of intra-system interactions from microcosm research we must simultaneously rely on mathematical models to extend our understanding of influences transmitted from one system to another (Likens & Bormann, 1974; Walter - Echols & Lichtenstein, 1977 & 1978).

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## LITERATURE CITED

- Anderegg, B. N., E. P. Lichtenstein, & J. D. Kemp. 1977. Effects of Lindane on DNA, RNA, and Protein Synthesis in Corn Roots. *J. Agric. Food Chem.* 25: 923-928.
- Anderson, R. V. & D. C. Coleman. 1977. The Use of Glass Microbeads in Ecological Experiments with Bacteriophagic Nematodes. *J. Nematology* 9: 319-322.
- Anderson, R. V. D. C. Coleman, C. V. Cole, E. T. Elliott. In Press. Effect of the Microbivorous Nematodes Actoboloides sp. and Mesodiplogaster lheritieri on Substrate Utilization and Nitrogen and Phosphorus Mineralization in Soil.
- Anderson, R. V., E. T. Elliott, J. F. McClellan, D. C. Coleman, C. V. Cole, & H. W. Hunt. 1978. Trophic Interactions in Soils as they Affect Energy and Nutrient Dynamics. III. Biotic Interactions of Bacteria, Amoeba, and Nematodes. *Microbial Ecology* 4: 361-371.
- Anderson, R. V., D. C. Coleman, C. V. Cole, E. T. Elliott, & J. F. McClellan. 1979. The Use of Soil Microcosms in Evaluating Bacteriophagic Nematode Response to Other Organisms and Effects on Nutrient Cycling. *Intern. J. Environmental Studies* 13: 175-182.
- Anonymous. 1976. Microcosm Research. *Bull. Ecol. Soc. Amer.* 57: 15.
- Atlas, R. M., D. Pramer, & R. Bartha. 1978. Assessment of Pesticide Effects on Non-Target Soil Microorganisms. *Soil Biol. Biochem.* 10: 231-239.
- Ausmus, B. S., D. R. Jackson, P. Van Voris, G. J. Dodson, E. G. O'Neill, B. M. Ross. 1977. The Use of Microcosms in Terrestrial Ecosystem Analysis. (Abstr.) *Bull. Ecol. Soc. Amer.* 58: 48.
- Ausmus, B. S., G. J. Dodson, & D. R. Jackson. 1978. Behavior of Heavy Metals in Forest Microcosms: III. Effects on Litter - Soil Carbon Metabolism. *Water, Air, & Soil Pollution* 10: 19-26.
- Ausmus, B. S., S. Kimbrough, D. R. Jackson, & S. Lindberg. 1979. The Behavior of Hexachlorobenzene in Pine Forest Microcosms. Transport and Effects on Soil Processes. *Environmental Pollution* : 103-111.
- Ausmus, B. S. & E. G. O'Neill. 1977. Comparison of Carbon Dynamics of Three Microcosm Substrates. *Soil Biol. Biochem.* 10: 425-429.
- Bartha, R. & D. Pramer. 1965. Note: Features of a Flask and Method for Measuring the Persistence and Biological Effects of Pesticides in the Soil. *Soil Sci.* 100: 68-70.
- Beall, M. L., Jr., R. G. Nash, & P. C. Kearney. 1976. Agroecosystem - A Laboratory Model Ecosystem to Simulate Agricultural Field Conditions for Monitoring Pesticides. IN: EPA 600/9-76-016, July 1976. Proceedings of the Conference on Environmental Modeling and Simulation.

- Bond, H., R. Lighthart, R. Shimabuku, & L. Russel. 1976. Some Effects of Cadmium on Coniferous Forest Soil and Litter Microcosms. *Soil Sci.* 121: 278-287.
- Bond, H., R. Lighthart, & R. G. Volk. 1977. The Use of Soil/Litter Microcosms With and Without Added Pollutants to Study Certain Components of the Decomposer Community. pp. 111-121. IN: M. Witt & J. W. Gillett, eds. *Terrestrial Microcosms and Environmental Chemistry*. The Proceedings of two colloquia held at Oregon State University as a part of the Symposium on Terrestrial Microcosms and Environmental Chemistry, June 13-14, 1977.
- Bondietti, E. A., S. Draggan, F. B. Taub, N. Pearson, J. R. Trabalka, & J. P. Wither-  
spoon. 1976. State - of - the - Art and Proposed Testing for Environmental  
Transport of Toxic Substances. ORNL, Environmental Sciences Div., Oak Ridge,  
TN 37830.
- Coats, J. R., R. L. Metcalf, P - Y. Lu, D. D. Brown, J. F. Williams, & L. G. Hansen.  
1976. Model Ecosystem Evaluation of the Environmental Impacts of the Veterinary  
Drugs Phenothiazine, Sulfamethiazine, Clopidol, and Diethylstilbestrol.  
*Environm. Health Perspectives* 18: 167-179.
- Cole, C. V., E. T. Elliott, H. W. Hunt, & D. C. Coleman. 1978. Trophic Inter-  
actions in Soils as they Affect Energy and Nutrient Dynamics. V. Phosphorus  
Transformations. *Microbial Ecology* 4: 381-387.
- Cole, L. K., R. L. Metcalf. 1977. Predictive Environmental Toxicology of Pesticides  
in the Air, Soil, Water and Biota of Terrestrial Model Ecosystems. IN: J. M.  
Witt & J. W. Gillett, eds. *Terrestrial Microcosms and Environmental Chemistry*.
- Cole, L. K., J. R. Sanborn, & R. L. Metcalf. 1976 a. Inhibition of Corn Growth  
by Aldrin and the Insecticide's Fate in the Soil, Air, Crop, and Wildlife of a  
Terrestrial Model Ecosystem. *Environm. Entomol.* 5: 583-589.
- Cole, L. K., R. L. Metcalf, & J. R. Sanborn. 1976 b. Environmental Fate of  
Insecticides in Terrestrial Model Ecosystems. *Intern. J. Environm. Studies*  
10: 7-14.
- Coleman, D. C., C. V. Cole, R. V. Anderson, M. Blaha, M. K. Champion, M. Clarholm,  
E. T. Elliott, H. W. Hunt, B. Shaefer, & J. Sinclair. 1977. An Analysis of  
Rhizosphere - Saprophyte Interactions in Terrestrial Ecosystems. *Ecol. Bull.*  
(Stockholm) 25: 299-309.
- Coleman, D. C., R. V. Anderson, C. V. Cole, E. T. Elliott, L. Woods, & M. K.  
Champion. 1978 a. Trophic Interactions in Soils as they Affect Energy and  
Nutrient Dynamics. IV. Flows of Metabolic and Biomass Carbon. *Microbial  
Ecology* 4: 373-380.
- Coleman, D. C., C. V. Cole, H. W. Hunt, and D. A Klein. 1978 b. Trophic Inter-  
actions in Soils as they Affect Energy and Nutrient Dynamics. I Introduction.  
*Microbial Ecology* 4: 345-349.

- Draggan, S. 1976 a. Microprobe Analysis of  $^{60}\text{Co}$  Uptake in Sand Microcosm. *Xenobiotica* 6: 557-563.
- Draggan, S. 1976 b. The Role of Microcosms in Ecological Research. *Intern. J. Environm. Studies* 10: 1-2.
- Draggan, S. 1976 c. The Microcosms as a Tool for Estimation of Environmental Transport of Toxic Materials. *Intern. J. Environm. Studies* 10: 65-70.
- Draggan, S. & J. M. Giddings. 1978. Testing Toxic Substances for Protection of the Environment. *The Science of the Total Environment* 9: 63-74.
- Draggan, S. & P. Van Voris. 1979. The Role of Microcosms in Ecological Research. *Intern. J. Environm. Studies* 13: 83-85.
- Elliott, E. T., C. V. Cole, D. C. Coleman, R. V. Anderson, H. W. Hunt, & J. F. McClellan. 1979. Amoebal Growth in Soil Microcosms: A Model System of C, N, and P Trophic Dynamics. *Intern. J. Environm. Studies* 13: 169-174.
- Eschenroeder, A., K. Tran, & C. Tashima. 1977. Mathematical Modeling of Toxic Substances in the Environment. *IN*: J. M. Witt & J. W. Gillett, eds. *Terrestrial Microcosms and Environmental Chemistry*. Proc. of two Colloquia held at Corvallis, Oregon, June 13-14, 1977.
- Fuhremann, T. W. & E. P. Lichtenstein. 1978. Release of Soil - Bound Methyl C - 14 Parathion Residues and Their Uptake by Earthworms and Oat Plants. *J. Agric. Food Chem.* 26: 605-610.
- Giddings, J. M. & G. K. Eddleman. 1977. The Effects of Microcosm Size and Substrate Type on Aquatic Microcosm Behavior and Arsenic Transport. *Arch. Environm. Contam. Toxicol.* 6: 491-505.
- Gile, J. D. & J. W. Gillett. 1979 a. Fate of Selected Fungicides in a Terrestrial Laboratory Ecosystem. *J. Agric. Food Chem.* 27: 1159-1164.
- Gile, J. D. & J. W. Gillett. 1979 b. Fate of C - 14 Dieldrin in a Simulated Terrestrial Laboratory Ecosystem. *Arch. Environm. Contam. Toxicol.* 8: 107-124.
- Gillett, J. W. & J. D. Gile. 1976. Pesticide Fate in Terrestrial Laboratory Ecosystems. *Intern. J. Environm. Studies* 10: 15-22.
- Gillett, J. W., J. Hill, IV, A. W. Jarvinson, & W. P. Schoor. 1974. A Conceptual Model for the Movement of Pesticides Through the Environment. EPA 600/e-74-024.
- Gillett, J. W. & J. M. Witt, eds. 1977. *Terrestrial Microcosms*. Proceedings of the Workshop on Terrestrial Microcosms, *Symposium on Terrestrial Microcosms and Environmental Chemistry*. National Science Foundation, Directorate for Applied - Science and Research Applications, Division of Problem - Focused Research Applications. Held at Corvallis, Oregon, Oregon State Univ., June 13-14, 1977.

- Gordon, R. W., R. J. Beyers, E. P. Odum, & R. G. Eagon. 1969. Studies of a Simple Laboratory Microecosystem: Bacterial Activities in a Heterotrophic Succession. *Ecology* 50: 86-100.
- Herrera, R. T., N. Merida, & C. F. Jordon. 1978. Direct Phosphorus Transfer from Leaf Litter to Roots. *Naturwissenschaftler* 65: 208-209.
- Herzberg, M. A., D. A. Klein, & D. C. Coleman. 1978. Trophic Interactions in Soils as they affect Energy and Nutrient Dynamics. II. Physiological Responses of Selected Rhizosphere Bacteria. *Microbial Ecology* 4: 351-359.
- Hirwe, A. S., R. L. Metcalf, P-Y. Lu, & L-C. Chio. 1975. Comparative Metabolism of 1,1 - bis - (p Chlorophenyl) - 2 - Nitropropane (Prolan) in Mouse, Insects, and in a Model Ecosystem. *Pesticide Biochem. & Physiol.* 5: 65-72.
- Jackson, D. R., C. D. Washburn, & B. S. Ausmus. 1977. Loss of Ca and NO<sub>3</sub> - N from Terrestrial Microcosms as an Indicator of Soil Pottution. *Water, Air, & Soil Pollut.* 8: 279-284.
- Jackson, D. R. & J. M. Hall. 1978. Extraction of Nutrients from Intact Soil Cores to Assiss the Impact of Chemical Toxicants on Soil. *Pedo - biologia* 18: 272-278.
- Jackson, D. R., W. J. Selvidge & B. S. Ausmus. 1978 a. Behavior of Heavy Metals in Forest Microcosms: I. Transport and Distribution Among Components. *Water, Air, & Soil Pollut.* 10: 3-12.
- Jackson, D. R., & B. S. Ausmus. 1978 b. Behavior of Heavy Metals in Forest Microcosms: II. Effects on Nutrient Cycling Processes. *Water, Air, & Soil Pollut.* 10: 13-18.
- Jackson, D. R., B. S. Ausmus, & M. Levin. 1979. Effects of Arsenic on Nutrient Dynamics of Grassland Microcosms and Field Plots. *Water, Air, & Soil Pollut.* 11: 13-21.
- Jackson, D. R. & M. Levin. 1979. Transport of Arsenic in Grassland Microcosms and Field Plots. *Water, Air, & Soil Pollut.* 11: 3-12.
- Kapoor, I. P., R. L. Metcalf R. F. Nystrom, & G. K. Sangha. 1970. Comparative Metabolism of Methoxychlor, Methiochlor, and DDT in Mouse, Insects, and in a Model Ecosystem. *J. Agric. Food Chem.* 18: 1145-1152.
- Kapoor, I. P., R. L. Metcalf, A. S. Hirwe, P-Y. Lu, J. R. Coats, & R. F. Nystrom. 1972. Comparateve Metabolism of DDT, Metabolism of DDT, Methylchlor, and Ethoxychlor in Mouse, Insects, and in a Model Ecosystem. *J. Agric. Food Chem.* 20: 1-6.
- Kaufman, D. D. 1977. Biodegradation and Persistence of Several Acetamide, Acetanilide Azide, Carbamate, and Organophosphate Pesticide Combinations. *Soil Biol. Biochem.* 9: 49-57.

- Klein, D. A. 1977. Seasonal Carbon Flow and Decomposer Parameter Relationships in a Semiarid Grassland Soil. *Ecology* 58: 184-190.
- Klein, D. A., P. A. Mayeux, S. L. Seaman. 1972. A Simplified Unit for Evaluation of Soil Core Respirometric Activity. *Plant & Soil* 36: 177-183.
- Klein, W., M. Anagnostopoulos, S. Begum, E. Elsner, D. Freitag, S. Cab, W. Greb, A. Hague, K. Hustert, L. Kilser, J. Kohli, F. Korte, D. Kotzias, J. P. Lay, R. N. Moza, H. Muller, W. Muller, W. Muller, G. Nohynek, A. Parlar, D. Prestel, K. Sandrock, N. Sotiriou, S. Viswanathan, D. Vockel, & I. Weisgerber. 1975. Comparative Behavior of Organochlorine Insecticides and Related Chemical Residues in Model Ecosystems. pp. 35-60. IN: Origin and Fate of Chemical Residues in Food, Agriculture, and Fisheries. Proc. and Report of Two Research Coordination Meetings held in Vienna, Austria, Nov. 5-9, 1973, and June 4-7, 1974. Organized by the Joint FAO / IAEA Division of Atomic Energy in Foods and Agriculture.
- Kudeyarov, V. N. & D. S. Jenkinson. 1976. The Effect of Biocidal Treatments on Metabolism in Soil - VI: Fumigation with Carbon Disulphide. *Soil Biol. & Biochem.* 8: 375-378.
- Kuntsman, J. L. & E. P. Lichtenstein. 1979. Effects of Nutrient Deficiencies In Corn Plants on the in Vivo and in Vitro Metabolism of C - 14 Diazinon. *J. Agric. Food Chem.* 27: 770-774.
- Lichtenstein, E. P. 1977. Fate of Pesticides in a Soil - Plant Microcosm. pp. 95-101. IN: J. M. Witt & J. W. Gillett, eds. Terrestrial Microcosms and Environmental Chemistry. NSF.
- Lichtenstein, E. P., T. W. Fuhremann, K. R. Schulz, & R. F. Skrentny. 1967. Effect of Detergents and Inorganic Salts in Water on the Persistence and Movement of Insecticides in Soils. *J. Econ. Entomol.* 50: 1714-1721.
- Lichtenstein, E. P., T. T. Liang, & T. W. Fuhremann. 1978. A Compartmentalized Microcosm for Studying the Fate of Chemicals in the Environment. *J. Agric. Food Chem.* 26: 948-953.
- Lichtenstein, E. P., K. R. Schulz, & T. T. Liang. 1977. Fate of Fresh and Aged Soil Residues of the Insecticide C - 14 N - 2596 in a Soil - Corn - Water Ecosystem. *J. Econ. Entomol.* 70: 169-175.
- Lighthart, B., H. Bond, & M. Ricard. 1977. Trace Element Research Using Coniferous Forest Soil / Litter Microcosms. EPA 600 / 3 - 77 - 091.
- Lighthart, G., & H. Bond. 1976. Design and Preliminary Results from a Soil / Litter Microcosms. *Intern. J. Environm. Studies* 10: 51-58.
- Likens, G. E. & F. H. Bormann. 1974. Linkages Between Terrestrial and Aquatic Ecosystems. *Bioscience* 24: 447-456.
- Lu, P-Y, R. L. Metcalf, A. S. Hirwe, & J. W. Williams. 1975. Evaluation of Environmental Distribution and Fate of Hexachlorocyclopentadiene, Chlordene, Heptachlor, and Heptachlor Epoxide in a Laboratory Model Ecosystem. *J. Agric. Food Chem.* 23: 967-973.

- Lu, P-Y., R. L. Metcalf, N. Plummer, & D. Mandel. 1977. The Environmental Fate of Three Carcinogens: Benzo - (a)-Pyrene, Benzidine, and Vinyl - Chloride Evaluated in Laboratory Model Ecosystems. *Arch. Environm. Contamin. Toxicol.* 6: 129-142.
- Lugo, A. E. 1969. Energy, Water and Carbon Budgets of a Granite Outcrop Community. Ph.D. Dissertation, Botany. University of North Carolina, Chapel Hill.
- McCormick, F. R., A. E. Lugo, & R. R. Sharitz. 1974. Experimental Analysis of Ecosystems, pp. 150-179. IN: B. R. Strain & W. D. Billings, eds. Handbook of Vegetation Science. Part VI: Vegetation and Environment. Dr. W. Junk, B. V., the Hague.
- McCormick, J. F. & R. B. Platt. 1962. Effects of Ionizing Radiation on a Natural Plant Community. *Rad. Bot.* 2: 161-188.
- Metcalf, R. L. 1974 a. Laboratory Model Ecosystem to Evaluate Compounds Producing Biological Magnification. *Essays Toxicol.* 5: 17-38.
- Metcalf, R. L. 1974 b. Development of Laboratory Model Ecosystem as Early Warning Elements of Environmental Pollution. Aerospace Medical Research Laboratory, Wright - Patterson AFB, Ohio. Report No. AMRL - TR - 74 - 125 - paper - 1; 12 pages.
- Metcalf, R. L. 1976. Laboratory Model Ecosystem Evaluation of the Chemical and Biological Behavior of Radiolabeled Micropollutants. *Environm. Qual. & Safety* 5: 141-151.
- Metcalf, R. L. 1977. Model Ecosystem Approach to Insecticide Degradation: A Critique. *Ann. Rev. Entomol.* 22: 241-261.
- Metcalf, R. L. & P-Y Lu. 1973. Environmental Distribution and Metabolic Fate of Key Industrial Pollutants and Pesticides in a Model Ecosystem. University of Illinois, Urbana - Champagne. Water Resources Center, Report No. 69.
- Metcalf, R. L., I. P. Kapoor, P-Y Lu, C. K. Schuth, & P. Sherman. 1973. Model Ecosystem Studies of the Environmental Fate of Six Organochlorine Pesticides. *Environm. Health Perspect.* June, 1973: 35-44.
- Metcalf, R. L., G. K. Sangha, & I. P. Kapoor. 1971. Model Ecosystem for the Evaluation of Pesticide Biodegradability and Ecological Magnification. *Env. Sci. & Tech.* 5: 709-713.
- Meyer, K. A. 1972. The Influence of Nutrient Availability and Nutrient Cycles upon Ecosystem Structure and Stability. Ph.D. Dissertation, Botany. University of North Carolina, Chapel Hill.
- Meyer, K. A., J. F. McCormick, & C. G. Wells. 1975. The Influence of Nutrient Availability on Ecosystem Structure. pp. 765-779. IN: F. G. Howell, J. B. Gentry & M. H. Smith, eds. Mineral Cycling in Southeastern Ecosystems ERDA Symposium Series CONF - 740513.

- Murphy, P. G. 1970. Ecological Effects of Acute Beta Irradiation from Simulated Fallout Particles upon a Natural Plant Community. Ph.D. Dissertation, Botany. University of North Carolina, Chapel Hill.
- Murphy, P. G. & J. F. McCormick. 1971. Ecological Effects of Acute Beta Irradiation from Simulated Fallout Particles on a Natural Plant Community. IN: D. W. Benson & A. H. Sparrow, eds. Survival of Food Crops and Livestock in the Event of a Nuclear War. AEC Symposium Series 24.
- Nabholz, J. V. 1978. Nutrient Budgets and Effects of Perturbation in Rock Outcrop Ecosystems. Ph.D. Dissertation, University of Georgia, Athens.
- Odum, E. P. 1971. Fundamentals of Ecology, 3rd ed. W. B. Saunders Co., Philadelphia. 574 & xvi pages.
- Odum, H. T. & A. Lugo. 1970. Metabolism of Forest Floor Microcosms. IN: H. T. Odum, ed. Tropical Rain Forest. pp. 135-156.
- Olson, R. K. 1975. The Effects of Acid Precipitation on Nutrient Cycling in Granite Outcrop Ecosystems. M. A. Thesis, Botany. University of North Carolina, Chapel Hill.
- O'Neill, R. V., B. S. Ausmus, D. R. Jackson, R. I. Van Hook, P. Van Voris, C. Washburn, & A. P. Watson. 1977. Monitoring Terrestrial Ecosystems by Analysis of Nutrient Export. Water, Air, & Soil Pollut. 8: 271-277.
- Patten, B. C. & M. Witkamp. 1967. Systems Analysis of  $^{134}\text{Cs}$  Kinetics in Terrestrial Microcosms. Ecology 48: 813-824.
- Platt, R. B. & J. F. McCormick. 1964. Manipulatable Terrestrial Ecosystems. Ecology 45: 649-650.
- Sanborn, J. R., R. L. Metcalf, W. N. Bruce, & P-Y Lu. 1976. The Fate of Chlordane and Toxaphene in a Terrestrial - Aquatic Model Ecosystem. Environm. Entomol. 5: 533-538.
- Sanders, W. M. III. 1976. EPA Needs in Microcosm Research. Intern. J. Environm. Studies 10: 3-5.
- Sharitz, R. R. & J. F. McCormick. 1973. Population Dynamics of Two Competing Annual Plant Species. Ecology 54: 723-740.
- Shirazi, M. A. 1979. Development of Scaling Criteria for Terrestrial Microcosms. EPA 600 / 3 - 79 - 017. Corvallis Environmental Research Laboratories, Office of Research & Development, U. S. EPA, Corvallis, Oregon.
- Snyder, J. M. & L. H. Wullenstein. 1973. Nitrogen Fixation on Granite Outcrop Pioneer Ecosystems. Bryologist 76: 196-199.

- Spalding, B. P. 1978. The Effect of Biocidal Treatments on Respiration and Enzymatic Activities of Douglass - fir Needle Litter. *Soil Biol. Biochem.* 10: 537-543.
- Timbers, G. E., H. P. Hocking, & S. S. Batth. 1971. A Microenvironment Cabinet for Toxicological Studies. *Canadian Entomol.* 103: 1347-1350.
- Tu, D. M. 1978. Effects of Pesticides on Acetylene Reduction and Microorganisms in a Sandy Loam. *Soil Biol. Biochem.* 10: 451-456.
- Van Voris, P., R. V. O'Neill, W. R. Emanuel, & H. H. Shugart. In Press. Functional Complexity and Ecosystem Stability.
- Vissner, S. A., M. Witkamp, & R. C. Dahlman. 1973. Flow of Microbially Fixed Nitrogen in a Model Ecosystem. *Plant & Soil* 38: 1-8.
- Walter - Echols, G. & E. P. Lichtenstein. 1978. Movement and Metabolism of C - 14 Phorate in a Flooded Soil System. *J. Agric. Food Chem.* 26: 599-604.
- Witkamp, M. 1976. Microcosm Experiments on Element Transfer. *Intern. J. Environm. Studies* 10: 59-63.
- Witkamp, M. & B. S. Ausmus. 1975. Effects of Tree Species, Temperature, and Soil on Transfer of Manganese - 54 from Litter to Roots in a Microcosm. IN: F. G. Howell, J. B. Gentry, & M. H. Smith, eds. Mineral Cycling in Southeastern Ecosystems. CONF - 740513. Tech. Info. Ctr., ORNL, Oak Ridge, TN pp. 694-699.
- Witkamp, M. & M. L. Frank. 1969. Cesium - 137 Kinetics in Terrestrial Microcosms. pp. 635-643. IN: D. J. Nelson & F. C. Evans, eds. Proceedings of the Second National Symposium of Radioecology. NTIS No. 67 - 5 - 3.
- Witkamp, M. & M. L. Frank. 1970. Effects of Temperature, Rainfall, and Fauna on Transfer of Cs - 137, K, Mg, and Mass in Consumer - Decomposer Microcosms. *Ecology* 51: 465-474.
- Witt, J. M. & J. W. Gillett, eds. 1977. Terrestrial Microcosms and Environmental Chemistry. Proceedings of two Colloquia as part of the Symposium on Terrestrial Microcosms and Environmental Chemistry held at Oregon State University, Corvallis, Oregon, June 13-14, 1977. NSF Pub. No. NSF / RA 79 - 0026.
- Zentmeyer, G. A. 1955. A Laboratory Method for Testing Soil Fungicides, with Phytophthora cinnamomi as a Test Organism. *Phytopathology* 45: 398-404.

LAKES AND MICROCOSMS: EXTENDING MICROCOSM DATA TO AQUATIC  
ECOSYSTEMS

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ABSTRACT

The introduction of significant quantities of toxic elements into our environment makes it increasingly important to understand the role these toxins play in disrupting natural ecosystems. Because it is not feasible to field test the toxic properties of such materials on a widespread basis without disrupting the very ecosystems we are trying to protect, it is urgent that methods be developed that permit the laboratory study of the ecological effects of toxins. Recent advances in the use of laboratory microcosms--dynamic representations of aquatic ecosystems--suggest that microcosms may be useful in testing the impacts of toxic elements on aquatic ecosystems. By designing microcosm tests that cover a wide range of ecological conditions, it should become possible to identify the consequences of widespread use of a chemical.

One limitation of microcosms is the difficulty in accurately modeling certain important ecosystem parameters such as fish productivity, water transparency, odor, and disease vectors. These "macro-variables" are often strongly influenced by a number of parameters--so-called "micro-variables"--that can be directly investigated in microcosms. By analyzing in microcosms the effects of perturbations on micro-variables, and by using field data on the correlations between these micro- and macro-variables, it is possible to deduce information on how a macro-variable may react to a given environmental perturbation. This paper reviews some of the relevant ecological relationships and presents some significant correlations between microcosm parameters and factors of interest to society. The current limitations and potential of our ability to extract information from these relationships are also discussed.

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I. INTRODUCTION

Among the components of aquatic ecosystems, fish yield and productivity, water transparency, odor, and pathogens and their vectors, are of particular concern to society. Human activities often adversely affect these "macro-variables", and it would be useful to be able to anticipate the consequences of such activities. Although macro-variables do not lend themselves to laboratory investigation for reasons discussed below, they are often strongly influenced by a number of "micro-variables" that are amenable to laboratory study. These micro-variables include temperature, the composition and productivity of planktonic communities, the chemical composition of the water, and microbial decomposition activity. This paper will describe the limitations and potential of our ability to extract results about macro-variables from information on micro-variables obtained from laboratory microcosm studies. Also discussed are some of the complications inherent in excluding components from microcosms and the difficulties resulting from inadequate field information.

Microcosms offer a means by which chemical concentrations, species compositions, trophic structures, and physical characteristics such as light levels and temperature can be altered under controlled conditions. Microcosms, however, have some serious limitations. Some ecosystem components, such as fish, cannot be satisfactorily modeled in laboratory microcosms due to distorting influences (Jassby et al. 1977, Harte et al. 1979). Yet excluding these variables removes ecological forces that act upon other parts of aquatic systems. These constraints restrict the amount of reliable information that one can obtain directly from microcosms. The presence of fish, for example, distorts nutrient flows in small microcosms, while their absence also causes distortions by removing influences such as grazing pressure on zooplankton communities. Transparency is difficult to measure in laboratory systems due to the distortions introduced by the compression of a large water column into a relatively shallow container. Another macro-variable, odor, also cannot be readily analyzed in microcosms. Investigation of changes in odor generally requires long-term studies, which, in microcosms, tend to result in laboratory conditioning of biota, surface growth problems and a general divergence of the dynamics of microcosms away from those of the natural systems being studied. The absence of a realistic atmosphere above a laboratory microcosm also impedes such an investigation by eliminating variables such as wind-induced turbulence, precipitation, and atmospheric deposition of nutrients.

By choosing to model the micro-variables that can be most accurately incorporated into microcosms, and by investigating field data on correlations between micro- and macro-variables, we can learn to extrapolate from laboratory studies to actual field situations. With the adoption of certain design and operating procedures, microcosms can become more valuable for the assessment of these links.

## II. MICROCOSM DYNAMICS

Microcosms are not capable of modeling all types of ecological phenomena and so no microcosm can be a completely satisfactory representation of an ecosystem. Their usefulness, therefore, will come from our ability to extend the partial information we get from laboratory studies into more integrated ecological analyses. This can only be done if we understand how microcosms differ from natural ecosystems and how the micro-variables affect and are affected by the macro-variables.

Macro-variables cannot be studied in microcosms for a variety of reasons that depend on the characteristics of the particular component. Macrofauna, for example, will distort nutrient flows in and out of the different trophic levels, exceed the natural ratios of fauna to lower trophic levels and distort the ratio of dissolved organic carbon to particulate organic carbon (Jassby et al. 1977). Fish, however, are known to regulate the size, species composition, and productivity of zooplankton populations, as well as carbon flux and nutrient regeneration times (Hrbacek et al. 1961, Winberg 1970, Wetzel 1975). Thus, excluding fish from microcosms also excludes their related influences. The interpretation of microcosm data must take this into account. It is possible to include fish and to attempt to incorporate into the analysis the distortions that occur due to their presence; however, following Harte et al. (1979) we take the approach of excluding macro-fauna in order to permit the dynamics of microcosms to resemble more accurately natural systems.

The transparency of water is another macro-variable that is poorly modeled under laboratory conditions. The size of microcosms are such that a one-meter column of water in a laboratory may represent a column of water tens of meters deep in a lake. The scale distortions that arise from the

compression of that column are large enough to discourage much transparency analysis in microcosms. Microcosms large enough to model transparency are theoretically possible, however a significant sacrifice in simplicity of design and operation results as microcosm size is increased.

The difficulty in achieving accurate long-term tracking in microcosms prevents the study of environmental components such as odor that require long-term experiments. Surface growth problems and the eventual conditioning of biota significantly reduce the accuracy of microcosm behavior over time (Dudzic et al. 1979). Surface growth exerts a significant effect on microcosm metabolism within six or seven weeks of inoculation, and the biological communities in microcosms begin to deviate from communities in natural systems. At present, it is not known whether surface growth mitigation strategies (Harte et al. 1979) will permit reliable studies of odor in laboratory systems.

We are thus left with a series of micro-variables from which to deduce information about the dynamics of variables that cannot be directly studied. The question arises as to the limits on learning about the micro-variables themselves in microcosms. If macro-variables are not included, how reliable is micro-variable information from microcosms? The answer depends on the specific laboratory design and on the magnitude of the complicating effects that would be exerted by the macro-variables on the micro-variables. The latter issue is discussed in some detail in section IV. Microcosm designs will vary among different research groups. It is possible, however, to generalize about certain limits that appear to be common to aquatic microcosms. Over short periods, up to six or seven weeks, microcosms appear able to accurately replicate or track the dynamics of natural systems if care is taken in

their initial inoculation (Harte et al. 1979). In addition, the various planktonic predator-prey relationships exhibit a generic resemblance to natural systems. For longer periods, however, a number of problems arise that cause their resemblance to lakes to decrease. In particular, growth of periphyton becomes a problem and the conditioning of biota in the tanks reduces the tracking accuracy of the whole community. Solutions to these problems (such as periodic pouring to eliminate surface growth) are certainly possible, however, they tend to introduce other distortions that need to be analyzed in detail. Microcosms are thus somewhat limited in the extent of their applications, but work is in progress to further improve their design and operation.

### III. MICRO-VARIABLE EFFECTS ON MACRO-VARIABLES

The applicability of microcosms to impact assessment ultimately rests upon our ability to extend our interpretations of experimental studies on micro-variables to the macro-variables of interest that cannot be adequately modeled. Such interpretations can only come from the use of field data and correlation analyses that investigate the many links between micro-variables and macro-variables.

Determining how a macro-variable will respond to a perturbation of a micro-variable is a complicated problem. The net effect of a perturbation is determined by the magnitude and direction of many responses. It is apparent that to ignore a single response with a large magnitude can cause a misinterpretation of the net direction of the effect of an environmental stress. Obviously, in ecological analysis, it is a difficult task to isolate and measure all the effects of an environmental stress. An action that reduces the size of a zooplankton population, for example, may cause a decrease in the

population and productivity of fish that are dependent upon the zooplankton. The subsequent reduction in grazing upon zooplankton by the fish may permit the zooplankton population level to increase back to previously high levels. The net effect cannot be predicted without some understanding of the relative magnitude of the grazing pressure of the fish on the zooplankton, the strength of the relationship between zooplankton productivity and fish productivity, and the numerous other factors such as grazing pressure on the fish themselves that may play a role. The following sections will examine the principle responses of fish productivity and water transparency to changes in nutrient and chemical concentrations and planktonic species composition and population.

### III A. The Role of Water-Column Nutrient Composition

Changes in nutrient levels cause changes throughout freshwater ecosystems, although the multiplicity of pathways often obscures the direction of some of the effects. Increases in certain nutrient concentrations may cause increases in the productivity or standing crops of plankton and fish, decreases in transparency of water due to increases in biogenic turbidity, and alterations in the concentrations of other nutrient and chemical levels.

Ever since Liebig proposed the theory of the limiting nutrient around 1840, limnologists and other ecologists have attempted to apply the concept to natural situations where a multitude of complex interacting factors exist. It has often been assumed that if an effect is produced on a species in a carefully controlled situation, that effect will also be observed in the field. A chief source of confusion is the failure of researchers to incorporate the numerous parameters that, though not directly connected to an ecological variable, may have indirect effects. One example is the net effect of nutrient

enrichment on the biomass and productivities of different trophic levels. It has been observed that increases in nutrient levels can dramatically increase the biomass of benthic fauna and that systems in which nutrients are made abundant by artificial means will increase the size of some part of the standing crop of fish communities (see, for example, Warren et al. 1964). The complex interaction between increasing densities of plankton, changing species composition, increasing predation pressure from higher trophic levels, and shifting development of macrophytic vegetation make it difficult to predict the net effect of enrichment from microcosm analyses. With information about the effects on consumers at all levels, the problems of extrapolating from micro-variables to macro-variables will be greatly simplified.

A problem further complicating the interpretation of simple enrichment experiments is the direct release of nutrients by macrofauna, thus increasing the levels and cycling times of nutrients that are available to other trophic levels. Animals excrete or otherwise make available several nitrogenous compounds, phosphates and trace elements. It has been observed that living zooplankton nourish phytoplankton by the release of carbon dioxide in respiration. Sushtchenia (1958) demonstrated that algae increase photosynthetic activity in the presence of zooplankton. These effects can now be carefully studied in microcosm. Similarly, Backiel and LeCren (1967) described the stimulation of primary productivity by the more rapid turnover of essential nutrients resulting from egestion and excretion of fish. While the net effect of zooplankton grazing is to reduce the standing crop of algae, the regeneration of nutrients by macrofauna will permit the growth of additional algae that may not have occurred if the original crop had not been grazed. It is therefore possible that the total production of the primary producers will be greater than might otherwise have taken place if a dense, overcrowded

population had been permitted to develop.

A similar situation involves the nutrients lost from the water column through sedimentation. The regeneration of these nutrients positively affects the magnitude of primary production. The release of nutrients from the sediments is accomplished by the direct interaction between benthic organisms and the sediments and by physical processes that are indirectly controlled by the activities of organisms (Edmondson 1961). The significance of this for microcosm research varies with the system being studied. Microcosms that represent a column of water should be able to incorporate the slight effects of nutrient regeneration fairly easily due to the absence of sediments (Dudzik et al. 1979). Microcosms that include sediments and perhaps even benthic organisms may also be able to include the effects of nutrient resuspension and regeneration, although the analysis will be considerably more complicated.

Another factor that affects the role of nutrients in altering productivities or standing crops is the difference in absorptive abilities of plankton at differing nutrient concentrations. Phytoplankton appear to absorb phosphates and nitrates at rates that depend on the nutrient concentrations (Edmondson 1961). If this is true for phytoplankton in general, then the instantaneous concentration of nutrients limits the total productivity. Similarly, Mann (1969) has shown that the rate at which zooplankton assimilate prey changes with nutrient availability and the prey species. When food is readily available, copepods ingest large quantities and assimilate very little (Cushing and Vucetic 1963), while other zooplankton increase their assimilation rates and store food for reserves. Some zooplankton species appear to be able to absorb and store excess phosphorus when concentrations are high, using the stored nutrients when concentrations are low.

The development of predictive models based on the levels of various nutrient and chemical species has been pursued as new and more complete analyses of aquatic ecosystems appear. Increasingly numerous regression analyses are beginning to show direct correlations among a number of principle factors. Rawson (1952, 1955), Ryder (1978), and Ryder et al. (1974) were among the first to analyze the role of various edaphic factors, such as the geology and soil profile of watersheds, and the morphology of lakes as crucial abiotic influences on biotic variables such as productivity and nutrient and energy cycling. Similarly, various researchers have developed predictive measures that correlate nutrient levels (such as phosphorus concentrations) with standing crop of phytoplankton (Dillon and Rigler 1974) and with annual fish yield (Hrbacek 1969).

Perhaps the most carefully studied analysis directly related to the role of nutrients and chemical compositions on macro-variables is the morphoedaphic index (MEI) which, in its simplest form, is the ratio of total dissolved solids (TDS) to the mean depth of a lake. Rawson (1952) recognized that the depth of a lake is important for its effect on temperature, thermal stratification, and the circulation and dilution of nutrients, and he suggested that the mean depth of a lake may be correlated with such variables as the sustained fishing yield of a lake. He then provided data to support this hypothesis by regressing biomass curves on mean depth to show that they exhibit an inverse hyperbolic relationship similar to that for fish yield (Rawson 1955). Kemp (1971) observed that total dissolved solids may be a more important variable than the chemical composition in any practical classification of waters. Total dissolved solids has often been assumed to be proportional to one or more of its vital or limiting components such as phosphorus or nitrogen. As an indicator of fish yield, therefore, TDS may be more useful than

any of its single component ions that are more susceptible to temporal and spatial variation. In this respect, TDS bears a similar relation to nutrient loading (see Vollenweider 1969) as standing crop bears to actual fish production (Ryder et al. 1974). Other researchers have also used TDS and found it to be a good indicator of various ecological conditions. Northcote and Larken (1956) found TDS to be the best single environmental indicator of the general level of productivity in the 100 lakes of British Columbia that they studied. Similarly, Jenkins (1967) found TDS to be the best environmental correlate for the total standing crop of fish in United States reservoirs. Total dissolved solids can be readily measured in microcosms.

The morphoedaphic index (MEI) should be useful for estimating not only biomass or production of fish species, but also biomass and production of other biotic communities or any other component of the living or nonliving biotic segment (Ryder et al. 1974). It is important to find satisfactory methods for measuring these components and to determine other correlates that relate micro-variables to macro-variables. If such methods can be developed and satisfactory correlates identified, it should be possible to develop predictive models. Such models may permit microcosms to be used to assess the impact of a proposed activity. In particular, by using microcosms to determine the effect of a proposed activity on the level of total dissolved solids, or by altering TDS levels and studying the effects on organisms, it may be possible to use the MEI to evaluate the effect of the same activity on different lakes classified by various morphological characteristics. It should be noted that in natural systems where TDS is primarily affected by watershed activity, the use of microcosms to study TDS may not be appropriate.

The significance of good predictive models for enhancing the usefulness of microcosms is tremendous. If other, more accurate correlations such as those between nutrient concentrations and fish yield or general productivity can be made, it will be possible to extrapolate microcosm data to macro-variable responses without having to identify every intermediate consequence. This would greatly simplify the problems of accurately interpreting microcosm data.

Nutrient and other chemical concentrations also affect the transparency of water. Transparency or turbidity is a function of both biogenic and abiotic factors. The relative importance of each depends upon the nutrient and trophic conditions of the water and the nature of the soil in the surrounding watershed. Alterations in nutrient and chemical concentrations thus have consequences for the transparency of a body of water by affecting the composition and concentration of planktonic organisms and inorganic suspended material. In turn, the concentration of planktonic organisms and other biotic particulate matter determines the biogenic turbidity of a body of water. The species of plankton also plays a role in determining the color of the water when biogenic turbidity is high. Blue-green algae, for example, may cause surface waters to appear blue-green, while an abundance of diatoms may cause a yellowish-brown color (Wetzel 1975).

Biogenic turbidity may sometimes be the principal source of turbidity and in such circumstances measurements of transparency (such as Secchi disk values) can be used to determine algal biomasses, or more precisely, the number of algal particles (Brezonik 1978). In San Francisco Bay, for example, biogenic turbidity varies from between three and thirty percent of the turbidity maximum over the year (Conomos and Peterson 1974). Small and Curl (1968)

determined that the concentration of chlorophyll a in Oregon coastal water was the principal factor responsible for the extinction of light in all seasons except summer, at which time non-chlorophyllous detritus greatly affected light attenuation.

Specific nutrients have also been related to levels of primary productivity and to levels of turbidity from biogenic sources. The spring total phosphorus loading of a lake, for example, can be used to predict the average summer chlorophyll a concentration, which in turn can be used to estimate the mean Secchi disk visibility--a measure of the transparency conditions of a body of water (Dillon and Rigler 1975). Thus, by using microcosms to determine how phosphorous levels will react to a given perturbation, it is possible to predict how the transparency of a lake may react to the same perturbation. Conversely, by setting limits on acceptable summer transparency conditions, one can determine permissible phosphorous loading in a lake at spring overturn. Transparency conditions have also been directly correlated to the concentrations of chlorophyll in several Scandanavian lakes (Ryding and Forsberg 1976).

Care must be taken in interpreting this relationship where there may be significant abiogenic turbidity due to the existence of high concentrations of inorganic particulate matter. Juday and Birge (1933) concluded that color was more important than plankton levels in decreasing transparency in 470 lakes of northern Wisconsin. The presence of inorganic color in standing waters has important implications for the use of Secchi disk transparency standards to delineate trophic state classes. In lakes with high levels of inorganic color, transparency values normally found in highly eutrophic waters may actually arise in some non-eutrophic situations. In brown water lakes and other

bodies of water with high levels of clay turbidity, Secchi disk readings are lower than would be expected from levels of chlorophyll a alone (Conomos and Peterson 1974). For this reason it is important to consider levels of abiotic color when using transparency values to determine trophic conditions.

Identifying the factors that control both biogenic and abiogenic turbidity has important implications for microcosm research. Concentrations of planktonic organisms, often responsible for biogenic turbidity, can be satisfactorily modeled in laboratory microcosms and if measurements of transparency can be used to determine algal populations (Brezonik 1978), one ought to be able to estimate transparency values from measurable algal populations in microcosms. Particle size, cell transparency, volume, and area may play a role in the accuracy of such estimations, but these variables also appear amenable to microcosm study and it may be possible to obtain useful correlations. Similarly, the correlation observed by Dillon and Rigler (1975) between phosphorus loading of a lake and Secchi disk visibility indicates that it is possible to relate transparency conditions to specific nutrient concentrations. It should be noted that certain characteristics of microcosms may limit their use in specific cases. For example, microcosms that model only a small section of water column will not have significant diel migration of plankton observed in natural lakes, with the corresponding fluctuations in transparency. Similarly, microcosms that do not incorporate sediments are also excluding a source of abiogenic turbidity.

As better correlations between micro-variables and macro-variables are identified, the next step is to be able to predict adverse consequences to macro-variables arising from certain micro-variable conditions. One example of this type of application is the correlation determined by Barica (1975)

between late-winter ammonia nitrogen concentrations, summer chlorophyll maximum concentrations and subsequent summer fish kills in certain ponds in southwestern Manitoba, Canada. Barica was able to show that lakes with certain consistent morphologic characteristics were very likely to suffer summer fish kills if concentrations of ammonia nitrogen the previous winter were above a certain level. The ammonia nitrogen concentrations above a certain point lead to large quantities of algae, algal blooms, rapid algal cell decomposition, oxygen depletion, and fish kills. Microcosms are capable of accurately modeling the dynamic variations of ammonia nitrogen levels and can be used to investigate how ammonia nitrogen levels may fluctuate with a given environmental perturbation. The ability to predict severe fish kills by seeing how ammonia nitrogen concentrations vary under controlled circumstances is precisely the sort of role microcosms can play in environmental assessments. As predictive models are refined to a greater degree, the accuracy of using microcosms to trace the effects of micro-variable perturbations will increase.

### III B. The Role of Planktonic Variables

Surprising, there appears to have been somewhat less success in developing predictive models to assess the direct effects of changes in specific planktonic communities on the different macro-variables such as fish yield and transparency. While the factor that most directly affects fish yields and productivity is the availability of food, there are numerous complicating species-specific effects that make narrowly-focused predictive models difficult to formulate. Some of these complications are discussed below.

High fish production rates requires high rates of fish-food production, not simply a large standing population of food organisms. The availability of

food alone, however, does not guarantee high productivity of fish populations since numerous other factors influence productivity. At low concentrations of food, fish populations either die out or are reduced to low levels, and all available food is used for maintenance of the existing population. At higher food concentrations, greater quantities may be used for growth or reproduction. In a study on fish production, Warren and his associates (1964) showed that when fish biomass is small, the fraction of food used for growth is significantly greater than that used for maintenance. Conversely, when fish biomass is large, most of the available food is required for maintaining fish stocks, leaving little for growth. Clarke (1946) pointed out that the existence of a large standing crop at one trophic level implies the existence of either a large initial standing crop of the prey organism (or nutrient) on which it feeds, or a high productivity, or rate of production of this organism.

These complications suggest that more general relationships between variables representative of planktonic productivity (such as nutrient levels) and macrofauna production may be easier to develop than species-specific relationships. There are a number of reasons why this may be so. First, generation times of plankton are typically much shorter than those of fish. Thus, changes in specific plankton populations due to a perturbation may not be immediately observable as a change in fish population or yield due to complications such as food switching and other behavioral characteristics of fish. On the other hand, large nutrient or chemical perturbations or changes in overall plankton populations will be reflected by changes in higher trophic levels and this may mask the effects of small temporal or spatial changes in single nutrient levels or species populations. The usefulness of total dissolved solids as a more accurate measure of chemical composition than any of its single component

ions is one example. Variables that are less susceptible to temporal and spatial effects may prove to be more useful in developing predictive models.

Another such variable is gross photosynthesis measured as diel oxygen changes in unconfined waters. McConnell et al. (1977) determined that fish yield and gross photosynthesis of phytoplankton and attached plants were highly correlated for six ecosystems studied. Melack (1976) recently reviewed most fish yield-gross photosynthetic relationships and he described the numerous correlations and studies. More recently, a model has been developed to predict the nighttime decline in dissolved oxygen concentrations as a function of planktonic respiration, fish respiration, respiration of other organisms, and oxygen diffusion (Boyd et al. 1978). The calculated dissolved oxygen concentrations usually agree to within ten percent of the measured values. These relationships permit Secchi disk visibility to be used to estimate consumption of oxygen by planktonic communities in ponds where plankton are the primary source of turbidity.

The depression (or stimulation) of plankton productivities (by changes in oxygen availability, for example), will cause a depression (or stimulation) of fish productivities. The response will not necessarily be proportional to the magnitude of the perturbation. This is due to food-switching, changes in food-assimilation rates, and other factors discussed in greater detail above. Nevertheless, a significant disturbance will certainly appear in the higher trophic levels as a significant response.

An interesting and important point must be made about the case where no response to a perturbation is observed in a microcosm. It is possible, and often observed in natural systems, that toxic substances may not affect lower trophic levels, but that biomagnification effects may appear at a later date

in a higher part of the food chain. Unless one specifically traces the physical presence of a toxin in an organism that may show no outward evidence of its presence, it is possible to entirely miss the effects of its presence until effects in higher trophic levels appear. In cases such as these, incorrect conclusions may be drawn using microcosm data about the effects of certain types of environmental disruptions or disturbances.

#### IV. COMPLICATING FACTORS

The preceding two sections have discussed how specific macro-variables, notably fish productivity and populations, and transparency conditions, respond to changes in micro-variables. There are other factors, however, that also play a large role in influencing the dynamics of fish populations. Section IV A. describes the most important of these factors and discusses the role of microcosms in developing methods to incorporate them into predictive ecosystem models. The other major complication is the effect exerted by macro-variables on the micro-variables themselves. Section IV B. discusses these influences and their significance for interpreting microcosm data.

##### IV A. Other Influences on Fish

Fish productivities are strongly dependent upon nutrient and chemical concentrations and the availability of food organisms, as described above. But other factors also enhance or inhibit the productivity of fish, including water temperature, the density of the fish population itself, and rates of predation on the fish community.

Within the last decade, considerable research has been done on the relationship between production (P) and standing crop (biomass B) for many different aquatic organisms--the so-called P/B ratio. It has been observed that the P/B ratio for given groups of organisms is reasonably constant in unstressed aquatic ecosystems (Waters 1969, Jonasson 1972, Waters and Crawford 1973). Production has been considered for use as an indicator of the health of an ecosystem, assessing the impact of various environmental effects. Yet there has been little thorough analysis of the relationship between production of different trophic-levels, e.g. how the productivity of fish-food organisms can be correlated to the production or productivity of fish. If direct correlations can be made, it should become possible to trace the consequences of environmental impacts through an ecosystem to levels that are of direct interest to society, such as fish yields, transparency, odor, and disease transmission.

Recently, the use of regression analysis has shown that fish yields, phytoplankton standing crops and various morphoedaphic factors such as mean depth of a body of water, total dissolved solids, and soil conditions in a watershed, can be related within reasonable confidence limits (Oglesby 1977). A number of these components, particularly planktonic variables and total dissolved solids, can be simulated in microcosms. The strong correlation of these data is encouraging, though much more work needs to be done before such regressions can be used in environmental assessment.

#### IV A.1. Temperature

Fish production has been shown to be highly dependent on the temperature of the water. Maximum fish-feeding rates occur at a particular (species-specific) temperature and feeding drops off as the temperature varies from this level. Assimilation of food into tissue also generally drops off with any change from the optimal temperature. The optimal temperature itself varies with the age and species of fish, and tolerances to changes in temperature vary with age, size, and species. Another complicating factor is acclimatization of fish metabolism to temperature over a period of time. Krogh (1916) describes the variation of metabolic rate as a function of temperature and Winberg (1971) suggests the application of "temperature corrections" to production estimates to account for the effect of temperature on development and growth patterns. Temperature is easily controlled in microcosms and the effects of temperature changes can be extensively studied.

#### IV A.2. Biomass/Standing Crop

Another factor affecting fish production is the size and composition of the fish population itself. As described above, the size of the standing crop of fish populations is a limiting factor in how efficiently the available food is cropped. Furthermore, feeding habits are species-dependent and there is evidence that some species decrease individual consumption rates when populations are large, while others increase consumption due to the ability of large schools of fish to efficiently graze concentrations of plankton (Wetzel 1975).

#### IV A.3. Predation

Several studies have shown that predation (e.g. through fishing pressures) on fish populations can result in increased productivity and fish yields. Differences in natural lifespan, growth rates and age distribution will be important in determining how maximum yields may be obtained (Ricker 1948, Beverton and Holt 1957, Cushing 1975). Jensen (1976) noted that as fishing effort increases, fish yield increases to a maximum level and then begins to decline as the number of older and larger individuals decreases. Factors such as these cannot be measured or modeled in microcosms, but by using growth data, age distributions and other information on life cycles of fish, they can be considered when interpreting microcosm data.

#### IV B. Effects of Macro-variables on Micro-variables

The effectiveness of using microcosms for environmental assessment is partly compromised by the necessity of excluding various macro-variables. The size of microcosms restricts the types of organisms that may be included and the exclusion of these variables results in the elimination of certain influences that may play important roles in natural ecosystems.

This presents an apparent contradiction: the elimination of certain variables from microcosms may increase their resemblance to natural systems, yet in natural waters these macro-variables play significant roles. Is it thus valid to assume that the driving forces behind the dynamics of aquatic ecosystems are those that are included in microcosms and that the macro-variables separately play secondary roles? If this is true, does it necessarily follow that one can ignore the combined effects of the macro-variables without drawing incorrect conclusions about the dynamics of natural systems?

The answer depends upon the magnitude of the effects of the excluded variables. It is apparent that certain effects that are insignificant at one stage in an ecosystem cycle may later play a dominant role. Similarly, synergistic or antagonistic relationships may be obscured by an analysis of single-variable effects, and a multitude of small relationships may combine to overwhelm single, more obvious responses. As a result, if certain variables must be excluded from microcosms, it is crucial to know how their absence may alter the results obtained.

The following compilation of complicating factors is, of course, incomplete due to the extensive and often subtle relationships that exist in aquatic systems. It does present some of the greatest effects that must be considered when analyzing dynamic responses to an environmental stress.

#### IV B.1. Effects of Fish

Fish exert a number of forces on planktonic communities that alter productivities, population levels, species composition, and size distributions. Perhaps the most important of these is the role of predation in altering the productivity and standing crop of the trophic level of prey organisms. The predation of organisms of one trophic level, such as fish, on the food organisms of lower trophic levels, such as zooplankton, tends to alter the size-distribution and species-composition of the standing crop, reduce the overall biomass, and increase the metabolism of the whole prey association (Hrbacek et al. 1961, Straskraba 1965). Predation effects are further complicated by apparent food-switching, age- and size-dependent food preferences, prey visibility, and food-assimilation rates that change with the population-density of the predators themselves.

Hrbacek and his fellow workers demonstrated the importance of fish predation in regulating the size and species composition of zooplankton and phytoplankton (Hrbacek et al. 1961, Hrbacek 1962, Hrbacek and Novotna-Dvorakova 1965, Straskraba 1967). In a series of experiments, the removal of fish from the Poltruba Backwater resulted in the development of large cladocerans at the expense of the smaller species of cladocerans, a reduction in rotifer populations, a decrease in the number of phytoplankton species, and an increase in the average size of individual phytoplankton of a given species. Mechanisms that are responsible--at least in part--include the selective feeding of fish on larger species of zooplankton, the stimulation effect of fish-feeding on the metabolism of planktonic communities, and the shift in the population under predation toward individuals of breeding age due to elimination of the larger and typically older individuals.

Studies of the size of gill-rakers of planktivorous and non-planktivorous fish show that size-selection naturally occurs due to filtering abilities. This is true of both obligate planktivores and those species that feed facultatively on the plankton (Wetzel 1975). Brooks (1968) has shown that freshwater planktivorous fish "actively search for and visually select each plankter that they ingest" and that fish predation on large zooplankton species is greater than on smaller species.

Even this size-specific predation is complicated by species-specific food preferences by fish populations. Ivlev (1961) demonstrated that fish exhibit marked preferences for certain prey species. When preferred species are not available, total food intake may actually decline, even in the presence of other types of food. Such species-specific preferences of fish, combined with changes in plankton productivity due to predation rates, can make

interpretation of microcosm data extremely difficult.

Another force that fish exert on planktonic communities is to alter the magnitude and composition of nutrients that cycle through aquatic ecosystems. The role of fish in the cycling of nutrients is quite complex, though the magnitude of the flows in relation to overall nutrient flows is relatively small. Through egestion and excretion, fish increase nutrient cycling in both running and standing waters. Certain species, such as catfish and carp, increase the rate at which nutrients are regenerated from sediments by their rooting and feeding actions, which stir up and resuspend sediments. Jassby et al. (1977) have shown that nutrient flow distortions occur in small microcosms when fish are included and that nutrient cycles in microcosms that exclude fish more closely resemble natural aquatic cycles. It is possible that nutrient flows in some lakes are sufficiently altered by fish to cause poor tracking responses in microcosms that exclude macrofauna. In these situations, either microcosms will have to be designed to include sediments and macrofauna or analytic methods of analysis will have to be formulated to account for the effects of the fish.

#### IV B.2. Effects of Light

The transparency of water is controlled by both biogenic and abiogenic factors and changes in transparency values have direct effects on numerous ecological variables. Light plays a crucial role in determining the productivity and dynamics of aquatic ecosystems. As transparency values change with fluctuating color and plankton levels, light availability for photosynthesis and primary production will vary.

At high levels of turbidity the size of the trophogenic zone (in which photosynthetic production occurs) decreases. Thus, increasing turbidity may cause a significant decrease in primary production. If a significant portion of the turbidity is of biogenic origin, a negative feedback effect may be observed, with turbidity decreasing as primary production decreases, leading to an increase in light penetration and a corresponding recovery of production rates. Because microcosm water columns are unnaturally shallow, these effects will be distorted in laboratory studies.

The rapid attenuation of light transmission by dense growths of bacteria and phytoplankton is commonly observed in stratified lakes (Wetzel 1975). The intensity of light directly controls the growth of algae and the rate of photosynthesis, though species-specific responses are observed. As biogenic turbidity of the algal populations increases in more fertile water, the productivity per unit area of water column does not increase in direct proportion due to the decreasing transparency and reduced size of the effective trophogenic zone. High productivity also depresses the plant nutrient concentrations and increases pH levels (Conomos and Peterson 1974).

Large algal population densities may cause shading of submerged vegetation, which in turn may decrease the habitat diversity of the littoral segment. Decreased littoral habitat diversity leads to decreases in the diversity and quantity of littoral macroinvertebrates.

Sabaneeff (1956) pointed out that high levels of turbidity interfere with the feeding mechanisms of zooplankton. High turbidity levels have also been observed to diminish the reactive distances of fish that feed visually (Vinyard and O'Brien 1976).

## V. SUMMARY

The use of aquatic microcosms to predict environmental effects on natural ecosystems is expanding as laboratory methods and techniques are refined and as our understanding of the dynamics of ecosystems increases. By using microcosms to investigate the response of micro-variables to a perturbation, it is possible to identify the principal mechanisms by which certain macro-variables will react, and to predict what their reactions might be.

This paper reviews some of the variables that can be incorporated in microcosms and that are responsible for affecting fish productivities and yields, and the transparency or turbidity of water. These variables, nutrient compositions and various planktonic factors, can be modeled in microcosms and subjected to controlled environmental stresses. We discuss how their reactions can then be observed and extended to trophic levels or variables that must be excluded from microcosms due to distorting influences.

Some correlation analyses between various ecological variables are also described in an attempt to identify the major relationships that might be useful in extending microcosm data to natural systems. Among the most promising possibilities are correlations of phosphorus levels with fish yields and transparency conditions; of total dissolved solids with the standing crop of fish; and of chemical concentrations such as ammonia nitrogen levels with subsequent oxygen depletion and fish kills. These and other correlations that will no doubt be identified offer good opportunities for microcosm research and the extrapolation of data on micro-variables to macro-variables of interest to society.

Finally, several complicating ecological effects are described in order to identify some of the difficulties in going from laboratory data to field predictions. Obviously, extensive work both in laboratory operation and design and ecological field analysis is still required, but it is possible to identify variables that are likely to provide useful predictive information when analyzed under controlled laboratory conditions.

REFERENCES

- Allen K.R. 1942. Comparison of bottom faunas as sources of available fish food. Trans. Am. Fish. Soc. 72:275-283
- Backiel T. and E.D. LeCren. 1967. Some density relationships for fish population parameters. In S.Gerking (ed.) The Biological Basis of Freshwater Fish Production (Blackwell Scientific Publication) p. 261-293
- Ball, R. C. and D.W. Hayne. 1952. Effects of the removal of the fish population on the fish-food organisms of a lake. Ecology 33 1:41-48
- Barica J. 1975. Summerkill risk in prairie ponds and possibilities of prediction. J. Fish. Res. Board Can. 32 1283-1288
- Beverton R.J.H. and S.J. Holt. 1957. On the dynamics of exploited fish populations. Fish. Invest. Lon. Ser. 19 2:533 pp.
- Boyd C.E., Romaine R.P. and E. Johnston. 1978. Predicting early morning dissolved oxygen concentrations in channel catfish ponds. Trans. Am. Fish. Soc. 107:484-492
- Brehmer M.L., Ball R.C. and N.R. Kevern. 1969. Nutrients and primary production in a warm-water stream. Department of Fisheries and Wildlife, Michigan State University (September)
- Brezonik P.L. 1978. Effect of organic color and turbidity on Secchi disk transparency. J. Fish. Res. Board Can. 35 1410-1416
- Brooks J.L. 1968. The effects of prey size selection by lake planktivores. Syst. Zool., 17 273-291
- Brylinsky M. and K.H. Mann. 1973. An analysis of factors governing productivity in lakes and reservoirs. Limnol. Oceanogr. 18 1-14
- Chapman D.W. 1967. Production in fish populations. In S.Gerking (ed.) The Biological Basis of Freshwater Fish Production (Blackwell Scientific Publ.) pp. 3-29
- Clarke G.C. 1946. Dynamics of production in a marine area. Ecological Monographs 16 4: 322-335
- Conomos T.J. and D.H. Peterson. 1974. Biological and chemical aspects of the San Francisco Bay turbidity maximum. In Mem. Inst. Geol. Bassin Aquitaine 7:45-52
- Curl H. Jr. and L.F. Small. 1965. Variations in photosynthetic assimilation ratios in natural, marine phytoplankton communities. Limnol. Oceanogr. 10, Suppl.: R67-R73
- Cushing D.H. 1975. Marine Ecology and Fisheries. (Cambridge University Press)

- Cushing D.H. and T. Vucetic. 1963. Studies on a Calanus patch. III. The quantity of food eaten by a Calanus finmarchicus. J. Mar. Bio. Assoc. U.K. 43 pp. 349-371
- Dillon P.J. and F.H. Rigler. 1974. The phosphorus-chlorophyll relationship in lakes. Limnol. Oceanogr. 19 767-773
- Dillon P.J. and F.H. Rigler. 1975. A simple method for predicting the capacity of a lake for development based on lake trophic status. J. Fish. Res. Board Can. 32 1519-1531
- Draggan S. 1976. The microcosm as a tool for estimation of environmental transport of toxic material. Intern. J. Environmental Stud. 10 pp. 65-70
- Dudzik M., Harte J., Jassby A., Lapan E., Levy D. and J. Rees. 1979. Some considerations in the design of aquatic microcosms for plankton studies. Intern. J. Environmental Studies. 13 125-130
- Edmondson W.T. 1961. Secondary production and decomposition. Verh. Internat. Verein. Limnol. 14 316-339
- Gerking S.D. 1962. Production and food utilization in a population of bluegill sunfish. Ecological Monographs 32 1:31-78
- Glooschenko W.A., Curl H. Jr. and L.F. Small. 1972. Diel periodicity of chlorophyll a concentrations in Oregon coastal waters. J. Fish. Res. Board Can. 29 1253-1259
- Harris G.P. and J.N.A. Lott. 1973. Light intensity and photosynthetic rates in phytoplankton. J. Fish. Res. Board Can. 30 1771-1778
- Harte J., Levy D., Rees J. and E. Saegerbarth. 1979. Making microcosms an effective assessment tool. Lawrence Berkeley Laboratory LBL-7892 (May) to appear in J. Geisey (ed.) Proceedings of the Symposium on Microcosms in Ecological Research (Augusta, Georgia November 1978)
- Henderson H.F., Ryder R.A. and A.W. Kudhongania. 1973. Assessing fishery potentials of lakes and reservoirs. J. Fish. Res. Board Can. 30 2000-2009
- Hrbacek J. 1962. Species composition and the amount of the zooplankton in relation to the fish stock. Rozpravy Ceskosl. Akad. Ved., Rada Matem. Prir. Ved. 72 1-114
- Hrbacek J. 1969. Relations between some environmental parameters and the fish yield as a basis for a predictive model. Verh. Internat. Verein. Limnol. 17 1069-1081
- Hrbacek J., Dvorakova M., Korinek V., and L. Prochazkova. 1961. Demonstration of the effect of fish stock on the species composition of zooplankton and the intensity of metabolism of the whole plankton association. Verh. Internat. Verein. Limnol. 14 192-195

- Hrbacek J. and M. Dvorakova-Novotna. 1965. Plankton of four backwaters related to their size and fish stock. Rozpravy Ceskosl. Akad. Ved., Rada Matem. Prir. Ved. 75 1-65
- Hynes H.B.N. 1963. The Biology of Polluted Waters (Liverpool University Press)
- Hynes H.B.N. 1970. The Ecology of Running Waters (Liverpool University Press)
- Ivlev V.S. 1961. Experimental ecology of the feeding of fishes. (Translated from the Russian by Douglas Scott, Yale University, New Haven, Conn.)
- Jassby A.M., Dudzik M., Rees J., Lapan E., Levy D. and J.Harte. 1977. Trophic structure modifications by planktivorous fish in aquatic microcosms. Environmental Protection Agency EPA-600/7-77-096. 52 pp
- Jenkins R.M. 1967. The influence of some environmental factors on standing crop and harvest of fishes in U.S. reservoirs. pp.298-321 In Reservoir Fisheries Resources Symposium. Southern Div. Am. Fish. Soc., Athens, Georgia
- Jensen A.L. 1976. Relation between yield and fishing mortality for Ricker's yield equation. J. Fish. Res. Board Can. 33 275-277
- Jonasson P.M. 1972. Ecology and production of the profundal benthos in relation to phytoplankton in Lake Esrom. Oikos Suppl. 14: 148 pp.
- Juday C. and E.A. Birge. 1933. The transparency, the color, and the specific conductance of the lake waters of northeastern Wisconsin. Trans. Wis. Acad. Sci., Arts, Lett. 28:205-259
- Kemp P.H. 1971. Chemistry of natural waters-VI. Classification of waters. Water Res. 5 945-956 (cited in Ryder et al. 1974)
- Krogh A. 1916. Respiratory exchange of animals and man. (London) (cited in Mann 1967)
- LeCren E.D. 1965. Some factors regulating the size of populations of freshwater fish. Mitt. Internat. Verein. Limnol. 13 pp. 88-105
- Leonard J.W. 1942. Some observations on the winter feeding habits of Brook Trout fingerlings in relation to natural food organisms present. Trans. Am. Fish. Soc. 72:219-227
- Mann K.H. 1967. The cropping of the food supply. In S.Gerking (ed.) The Biological Basis of Freshwater Fish Production (Blackwell Scientific Publications) pp. 243-257
- Mann K.H. 1969. The dynamics of aquatic ecosystems. Advances in Ecological Research 6 1-81

- Matuszek J.E. 1978. Empirical predictions of fish yields of large North American lakes. Trans. Am. Fish. Soc. 107:385-394
- Melack J.M. 1976. Primary productivity and fish yields in tropical lakes. Trans. Am. Fish. Soc. 105:575-580
- McConnell W.J., Lewis S. and J.E. Olson. 1977. Gross photosynthesis as an estimator of potential fish production. Trans. Am. Fish. Soc. 106:417-423
- Northcote T.G. and P.A. Larkin. 1956. Indices of productivity in British Columbia lakes. J. Fish. Res. Board Can. 13 515-540
- Oglesby R.T. 1977. Relationships of fish yield to lake phytoplankton standing crop, production, and morphoedaphic factors. J. Fish. Res. Board Can. 34 2271-2279
- Rawson D.S. 1952. Mean depth and the fish production of large lakes. Ecology 33 513-521
- Rawson D.S. 1955. Morphometry as a dominant factor in the productivity of large lakes. Verh. Internat. Verein. Limnol. 12 164-175
- Ricker W.E. 1946. Production and utilization of fish populations. Ecological Monographs 16 4:374-391
- Ricker W.E. 1948. Method of estimating vital statistics of fish populations. Indiana University Publ. Sci. Serv. 15 101 pp.
- Ryder R.A. 1978. Fish yield assessment of large lakes and reservoirs - a prelude to management. In S. Gerking (ed.) Ecology of Freshwater Fish Production 2nd edition (Blackwell Scientific Publications) pp. 403-423
- Ryder R.A., Kerr S.R., Loftus K.H. and H.A. Regier. 1974. The morphoedaphic index, a fish yield estimator-review and evaluation. J. Fish. Res. Board Can. 31 663-688
- Ryding S. and C. Forsberg. 1976. Six polluted lakes: A preliminary evaluation of the treatment and recovery process. Ambio 1976 pp.151-156
- Sabaneeff P. 1956. Uber das Zooplankton der Weser. Ber. Limnol. Flusstn Freudenthal 7 28-42. (cited in Hynes 1970)
- Small L.F. and H. Curl Jr. 1968. The relative contribution of particulate chlorophyll and river tripton to the extinction of light off the coast of Oregon. Limnol. Oceanogr. 13 84-91
- Small L.F., Curl H. Jr. and W.A. Glooschenko. 1972. Estimates of primary production off Oregon using an improved chlorophyll-light technique. J. Fish. Res. Board Can. 29 1261-1267
- Small L.F., Curl H. Jr. and W.A. Glooschenko. 1972. Effects of solar radiation and upwelling on daily primary production off Oregon. J. Fish. Res. Board Can. 29 1269-1275

- Straskraba M. 1965. The effect of fish on the number of invertebrates in ponds and streams. Mitt. Internat. Verein. Limnol. 13 pp. 106-127 (August)
- Straskraba M. 1967. Quantitative study on the littoral zooplankton of the Poltruba Backwater with an attempt to disclose the effect of fish. Ceskosl. Akad. Ved., Rozpravy Rada MVD 77 1-34
- Sushtchenia L.M. 1958. The influence of zooplankton on the intensity of photosynthesis of pond and lake plankton. Trudy Biol. Stat. on L. Narojoe. 1:223-240 (cited in Edmondson 1961)
- Thomas W.H. 1970. Effect of nitrate and ammonia concentration on chlorophyll increases in natural tropical Pacific phytoplankton populations. Limnol. Oceanogr. 15 386-394
- Van Oosten J. 1945. Turbidity as a factor in the decline of Great Lakes fishes with special reference to Lake Erie. Trans. Am. Fish. Soc. 75:281-322
- Vinyard G.L. and W.J. O'Brien. 1976. Effects of light and turbidity on the reactive distance of bluegill. J. Fish. Res. Board Can. 33 2845-2849
- Vollenweider R.A. 1969. Moglichkeiten und Grenzen elementarer Modelle der Stoffbilanz von Seen. Arch. Hydrobiol. 66:1-36 (cited in Ryder 1974)
- Vollenweider R.A. and A. Nauwerck. 1961. Some observations on the <sup>14</sup>C method for measuring primary production. Verh. Internat. Verein. Limnol. 14 134-139
- Wallen I.E. 1951. The direct effect of turbidity on fishes. Bull. Okla. agric. mech. Coll., Biol. Ser. 48 2: 27 pp. (cited in Hynes 1970)
- Warren C.E., Wales J.H., Davis G.E. and P. Doudoroff. 1964. Trout production in an experimental stream enriched with sucrose. Journal of Wildlife Management 28 4:617-660
- Waters T.F. 1969. Production in natural stream systems. In The Stream Ecosystem: An AAAS Symposium Michigan State University Institute of Water Resources (December)
- Waters T.F. and G.W. Crawford. 1973. Annual production of a stream mayfly population: A comparison of methods. Limnol. Oceanogr. 18 286-296
- Wetzel R.G. 1975. Limnology (W.B. Saunders Co. New York. 1975)
- Winberg G.G. 1970. Some interim results of Soviet IBP investigations on lakes. In Z. Kajak and A. Hillbricht-Ilkowska (eds.) Productivity Problems of Freshwaters Warsaw PWN. Polish Scientific Publishers. pp.363-381
- Winberg G.G. 1971. Methods for the Estimation of Production of Aquatic Animals (Academic Press, New York and London. Translated from the Russian by Annie Duncan, Royal Holloway College, University of London)

Wisemar R.C., Richey J.E. and D.E. Spyridakis. 1977. The importance of allochthonous particulate carbon pathways in a subalpine lake. J. Fish. Res. Board Can. 34 1410-1418



OPTIMUM MICROCOSMS FOR LAKE ECOTOXICOLOGY

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## INTRODUCTION

The major advantage of microcosms in ecotoxicology is that they permit study of complex ecosystems under conditions that allow a greater opportunity for manipulation and control compared with field studies. In addition, the expense and environmental risk entailed by microcosm investigations are relatively small compared with field studies of toxic substance effects. However, problems inherent in laboratory microcosms, such as diminished physical dimensions leading to excessive surface-to-volume ratios, and the absence of linkages with a realistic environment external to the immediate system, may undermine their potential advantages. Microcosms can be a useful tool in ecotoxicology only to the extent that: i) their biological and chemical behavior, under both undisturbed and disturbed conditions, are similar to that of natural systems, ii) identically initiated systems replicate well, iii) the attainment of i) and ii) does not result in excessive complexity or cost. The degree to which inherent problems of microcosms reduce their usefulness, in the above sense, will depend on the manner in which microcosms are set up and used. This paper will review the major options for microcosm design, initiation, and operation, emphasizing the ways in which the choices among these options influence the usefulness of the system. Our attention is restricted here to laboratory, freshwater-lake microcosms, although many of the conclusions will apply to other types of artificially confined ecosystems.

## DESIGN

The primary design decision for lake-like microcosms are the shape and size of the container. The traditional use of rectangular aquaria has little justification. The surface-to-volume ratio for such shapes is greater than necessary, thus enhancing the problem of surface growth. In addition, the

presence of unnecessary edges and corners makes sampling more difficult. Cylindrical containers are a better choice. The strength and durability as well as the chemical inertness of high-density polyethylene warrants the selection of this container material for many purposes\*.

A number of considerations affect the choice of container size. The sizes available range over three or four orders of magnitude, starting with systems of only a few liters in volume. The extrapolation from microcosms to natural lakes generally involves a much greater extension in volume. Thus, the best strategy is to operate microcosms in such a manner that their behavior is relatively size-independent, for only in that way can one have reasonable hope that the systems will behave like natural lakes.

As discussed in more detail in the section on operating microcosms, the dependence of microcosm behavior on container size can be reduced by the use of strategies to control surface growth. Size-independence is only achievable up to a point, however, and in our current investigations we have seen that even with surface-growth mitigation the similarity of a microcosm to the natural lake from which it is initiated begins to be severely reduced beyond a few weeks after initiation if the microcosm volume is less than about 15 liters.

If the investigator wishes to include macrofauna such as fish in the microcosm, then that places a major constraint on allowable size, for reasons given in the following section; closed microcosms with macrofauna must well exceed  $1 \text{ m}^3$  in volume if their behavior is to be realistic. The amount of water needed for periodic monitoring of a closed system also places a lower limit on the size, for one does not want sampling losses to consume a large fraction of the system. Monitoring of chemical nutrients (N, P, C) and phytoplankton and zooplankton densities typically requires about a hundred ml. per system. If the sampling is carried out weekly, then a 4-month run would result in the

\* It is ideally suited for toxic metal studies, but may not be as suitable as glass for the study of certain hydrocarbons that will adhere to the material.

loss of about 2 liters, and thus a system no smaller than about 20 liters would be needed to keep sampling loss to 10% of microcosm volume. Evaporation also represents a water loss, but this can be made up by the addition of deionized water.

In addition to the question of the proper size and shape of the microcosm container itself, there is the question of the appropriate dimensions of the benthic materials in a microcosm. If lake-bottom sediments are simply placed over the entire bottom area of the microcosm container, then these sediment materials will exert an unnatural influence on the water column above. The reason for this is simply the shallow depth of the microcosm compared with the lake (Jassby et. al., 1977a; Dudzik et. al., 1979). In order to solve this problem in marine microcosms, benthic chambers have been designed that provide a reduced surface area of sediment in contact with the surrounding water (Perez et. al., 1977). The realism of the behavior seen in such systems appears to be enhanced by this design. Extension of this approach to freshwater microcosms has not been carried out\*.

## INITIATION

The major degree of freedom available for microcosm initiation, and the one that we will focus on exclusively, is the choice and concentration of the initial chemical and biotic species. We describe here the various ways in which lake-like microcosms can be initiated and discuss the optimum uses for each approach.

### Gnotobiotic Initiation

At one extreme is the gnotobiotic, or "cookbook", approach in which a well-defined chemical medium is inoculated with selected quantities of organisms taken from pure cultures. A leading practitioner of this approach is Taub (1971, 1974). One advantage of gnotobiotic initiation is that it greatly

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\* It should be noted that in stratified lakes, for part of the year there is relatively little benthic influence on the epilimnion. In toxicological studies confined to the epilimnion, the need for a benthic chamber may be obviated.

simplifies taxonomic monitoring, for the reason that one knows what one is looking for. A second advantage is that simply-conceptualized situations can be created (for example, two predator species competing for one prey species) and thus the opportunity exists for testing the many, and often elegant, hypotheses based on simplifying abstractions in ecology (such as the competitive exclusion principle). Finally, by insuring the presence of a particular species of interest in the microcosm, the action of a toxicant on that species can be ascertained.

Study of the action of toxicants on an organism in a gnotobiotic system has the disadvantage that the environs of that organism are quite different from those found in nature, or even in a more complex microcosm. There is lacking presently an adequate measure of the "stress" on an assemblage of species that are brought together by actions bearing little or no resemblance to natural succession. Because this stress may act synergistically with the stress of toxicants, its description is essential if the gnotobiotic approach is to be of benefit in ecotoxicology.

A number of community-level ecotoxicological problems are highly unsuited for study in gnotobiotic systems. For example, investigation of the effects of toxicants on mineralization activity requires a realistic detritus pool as well as a wide and representative mix of detritivores. This would be extremely difficult to achieve under gnotobiotic conditions. A final problem concerning gnotobiotic microcosms for ecotoxicology is that of obtaining organisms. Gnotobiotic initiation appears to offer the potential for a high degree of standardization, simply because the organisms investigated can be listed

precisely. But with few exceptions, stocks of identical planktonic organisms are not widely available and invariant in time.

#### Whole-water-sample Initiation

At the opposite extreme from gnotobiotic initiation is the method of confining entire water samples, or water and sediment samples, in containers. With this approach, generally very complex communities are included in the microcosm. The initial chemical and biotic composition is, of course, completely determined by the characteristics of the natural source, although perturbations can then be carried out in order to study the effects of changes in the biotic or chemical composition of the microcosm. Other parameters, such as light levels, water temperature, and amount of water mixing are also completely at the disposal of the investigator. This approach was first explored in some detail by Beyers (1965). Its major advantage for ecotoxicology is that it offers the possibility of providing systems that will respond to a toxicant in much the same way as would the natural system from which the microcosm was initiated. This potential realism may not be achievable in practice, however; exploration of the degree of similarity between natural lakes and microcosms derived from those lakes by whole-water sampling is the subject of current research (Harte et.al., 1979).

Artificially confined whole water samples may behave quite differently from the natural system from which they originated for a number of reasons. Most importantly, the distorted surface-to-volume ratio of the confined system is likely to lead to "wall effects". Excessive growth of periphyton on the surfaces of the container is a sink for nutrients, thus reducing the similarity between the water-column chemistry of the microcosm and the parent lake (Whittaker, 1961; Dudzik et.al., 1979; Jassby et.al., 1977). Fortunately this problem is remediable, as discussed in the "Operation" section of this paper.

Another source of possible divergence between microcosms and parent lakes is the presence of macrofauna. If macrofauna, such as fish, are included in microcosms that are smaller in volume than on the order of  $10\text{ m}^3$ , their presence is likely to lead to gross distortions of nutrient flows and ambient physico-chemical conditions such as the ratio of dissolved to particulate organic carbon (Jassby et. al. 1977b; Harte et. al., 1979). On the other hand, the absence of fish macrofauna can lead to less-than-natural grazing pressure on zooplankton, and subsequent distortions in algal composition and community properties.

Macrofauna illustrate another problem with whole-water-sample initiation - that of small numbers. Organisms may be present in the parent water body at such low density that, at worst, their inclusion or exclusion is random while, at best, large fractional discrepancies will exist in the initial numbers present. This may be true for species of zooplankton as well as for larger organisms and has been observed to be a cause of poor replicability among microcosms (Jassby et. al., 1977a).

A difficult and as-yet-unresolved problem concerns the benthos. If the whole-water-sample includes benthic materials, the presence of the benthos may exert an excessive influence on the water column of the microcosm. This is a direct consequence of the shallow depth of the microcosm compared with the lake. (Jassby, et. al., 1977a; Dudzik, et. al., 1979). A potential solution to this problem lies not in the initiation procedure but in the design of the benthic compartment in the microcosm and was discussed in the "Design" section of this paper.

With the above caveats in mind, whole-water-sample initiation offers the best hope of constructing microcosms in which the biotic and chemical parameters mimic or track well those in the specific parent water body. It is

natural to inquire at this point as to the benefit of achieving such realism. Could not microcosms which behave like lakes in a generic sense be just as useful for ecotoxicology? We believe the answer is no for the following reason. Subsequent to the initiation of a lake-like microcosm, a marked succession of changes in biotic composition typically occurs over the first weeks or months. Often, for example, particular phytoplankton species will undergo a population bloom or crash. Similar changes are taking place in lakes at many times of the year as a result of natural successional processes, and so the generic property of population blooms and crashes may be well encapsulated in microcosms. But knowledge of the particular groupings of species that are blooming or crashing can be of the utmost importance in ecotoxicology because the stresses on the system that are exerted by the toxicant act in combination with natural forces of change. Thus microcosms that cannot track correctly these changes but are only models of lakes in a generic sense are likely to provide grossly distorted information about effects of toxicants in any particular lake. This does not mean that every lake about which one desires information must be studied afresh, for toxicant effects found in one lake can, to some extent, be extrapolated to wide classes of lakes. This seemingly paradoxical situation can be summarized as follows: One can learn specific information and deduce generic information about effects of toxicants from studies with microcosms that are able to simulate specific lakes; but one generally cannot develop either specific or generic information from microcosms that simulate only generic lake properties. realistic

#### Semi-gnotobiotic Initiation

Between the two extremes of gnotobiotic and whole-water-sample initiation, there are intermediate approaches in which some aspects of the biotic and chemical composition of the microcosm are determined at the outset entirely

by the parent water body and others are fixed by the investigator. One such approach is an inoculation method, in which a relatively small lake sample is added to a microcosm tank containing a nutrient medium defined by the investigator (Maguire, 1971; Neill, 1975; Jassby et. al., 1977a). In this way, a high diversity of organisms can be studied in a selected chemical environment. Lakes of any trophic state can be constructed, whether eutrophic or oligotrophic, whether N-limited or P-limited, according to the interests of the investigator.

Because the volume of the inoculum is usually a small fraction of the volume of the container in this approach, the organisms present at initiation find themselves in relatively enriched chemical environs. Thus, plankton blooms typically take place shortly following initiation, suggesting that this initiation method is particularly useful for the study of springtime succession phenomena. Studies have demonstrated surprisingly good agreement between generic succession patterns observed in such microcosms and those found in certain classes of natural lakes (Jassby et.al., 1977a). Using this initiation procedure, realistic tracking of a lake by a microcosm initiated from that lake (in the absence of day-to-day agreement of chemical and biotic composition) cannot be expected.

Another semi-gnotobiotic initiation method consists of adding selected organisms to microcosms initiated by a whole-water-sample. In this way, effects of toxic substances on organisms not found in large quantities in the parent system can be determined with greater statistical accuracy. Of course, this comes at the expense of reduced realism, because the higher-than-natural density of the organisms is likely to introduce distorting factors in the experiment.

Based on scattered and incomplete evidence, the whole-water-sample

initiation method appears to produce microcosms that replicate the best, at least over time periods of several weeks following initiation (Harte and Levy, 1979; Harte et. al. 1979). A careful study of the reasons for occasional poor replication of microcosms initiated by any of the methods described above has not been carried out. Investigation of this issue over running times of a month or longer is particularly needed.

## OPERATION

In the operation of lake-like microcosms a large number of degrees of freedom are available. We discuss two in detail here - namely, the choice of hydraulically closed versus open systems, and the choice among various approaches to the coping with the problem of excessive surface growth of algae - and then review briefly other, less critical, choices.

### Chemostatic versus Hydrostatic Systems

Lake microcosms can be either closed or open, hydraulically. In a closed system the water may be internally agitated but its residence time in the microcosm container is determined solely by the rate of water loss by evaporation and sampling. In such systems, chemical and biotic concentrations are determined by the internal dynamics of the microcosm. Hydraulically open, or flow-through, systems permit (in principle) the establishment of predetermined constant chemical concentrations, including the concentration of a toxicant being tested. The water exiting such chemostats either can be filtered so that all the organisms present remain within the system, or it can flush out the smaller organisms, in which case the biotic concentration and rate of flow of the replacement water to some extent determine the biotic concentrations within the system.

A microcosm model of a segment of a stream clearly calls for flow-through operation. Less obvious is the most appropriate operation of lake-like

microcosms. Advantages of the chemostat set-up have been reviewed by Veldkamp (1977). Because nutrients need never be in short supply in a chemostat, high concentrations of organisms over long time periods can be achieved. In conjunction with gnotobiotic initiation, for which chemostats are best suited (Porcella, 1969), such systems can allow large and constant populations of particular organisms and are thus convenient for certain types of analyses such as the determination of limiting nutrients for algal species. In contrast, in closed gnotobiotic systems the concentrations of nutrients and organisms are rarely constant.

The realism of such flow-through systems can be questioned, however. In natural lakes, time variation in concentration of both organisms and nutrients is the rule, not the exception. The value of assessments of toxicant effects in systems lacking these variations is not established. Moreover, carrying out ecotoxicological investigations on organisms at the high densities customary in chemostats can produce results that reflect the stresses of high density as well as those of the toxicant. Lastly in this connection, we point out that closed microcosms initiated by either whole-water-samples or semi-gnotobiotically do not require initial nutrient levels in excess of those found in natural waters in order that the organisms display growth rates and densities characteristic of those natural waters (Jassby et. al., 1977a; Harte and Levy, 1979).

Toxicants added to a microcosm will tend to accumulate on the surfaces, at rates that depend on the nature of the toxicant, container material, and the chemical and biotic composition of the microcosm. In a closed system, this can present a problem for the study of those toxicants that leave the water column too rapidly for toxicological information to be obtained. A reputed advantage of flow-through systems is that they allow the creation of constant water-column concentrations of toxicant, which are then more convenient for

establishing dose-response relations. This has been an important justification for going to the added expense and complexity of chemostats. However, it must be realized that even in flow-through systems some toxicants will accumulate on the surfaces. These surface-bound substances can still exert a toxic effect on the biota including the plankton, within the system, and thus constant water-column concentrations do not allow the investigator to assume constant exposure. Moreover, even neglecting surface effects, concentrations of toxicant in the microenvironment of organisms are likely not to be constant even though water-column concentrations are (Grossbard, 1972)

An important property of freshwater lakes is the average time-constant characterizing the flow-through of water. For many lakes, this time constant is of the order of a year or longer. Thus, over a period of a few weeks or months during which a toxicological study might take place in a microcosm, the lake being modeled will exchange only a small fraction of its water. A flow-through system that was designed to maintain constant levels of pollutants would necessitate a serious distortion in the hydrological properties of the microcosm relative to that of the lake. This would undoubtedly affect the biological realism of any results.

If one's goal is to maintain constant levels of a toxicant in an aquatic microcosm, it is far easier and less destructive of the realism of the system to simply measure the toxicant concentration at regular intervals and replace any losses. However, such a goal should not be adopted blindly. In some cases, pollutant levels in nature peak and then decline as biological, chemical, and physical processes operate. The strong pulse of acid input to Adirondack lakes following the spring thaw is one example. Learning how to extrace sensible measures of does-response under conditions of variable pollution levels is likely to be a far more fruitful enterprise than sacrificing biological realism by creating systems with artificially constrained constant

chemical conditions. It is our judgment that, when all factors are considered, hydraulically closed systems have a distinct advantage over chemostats for lake ecotoxicology if whole-water-sample initiation is employed.

#### Dealing with Surface Growth

The growth of periphyton on the inner surfaces of microcosm containers greatly constrains the use of any type of aquatic microcosm in ecotoxicology. It restricts the use of microcosms either to the first several weeks following initiation, when surface growth is not yet appreciable, or to periods when the plankton are undergoing accentuated blooms or crashes and their influence on nutrient exchanges is dominating that exerted by growth on the surfaces. The phenomenon of surface growth can lead to water-column behavior in a microcosm that is strongly dependent on the size of the microcosm, thus shedding doubt on the validity or realism of results.

Avoidance of rectangular containers helps, as discussed in the design section, but more effective procedures are needed. Three possible solutions can be envisioned. First is biological control of surface growth. Our experiments with snails (Physa sp.) and South American Catfish (Plecostumus sp.) to control surface growth of algae in large (700 liter) microcosms proved ineffective (Jassby et. al., 1977a). Reproduction of these organisms could not keep pace initially with periphyton growth; the long-stranded algae were immune to grazing; and, most importantly, the excretion and death of the grazers themselves distorted water-column conditions to much the same degree as did the surface growth.

A second approach involves mechanical scraping of the surfaces at periodic intervals. Our limited experience with this approach suggests it, too, is ineffective, except possibly in small glass containers. The surface growth is a mixture of algae and bacteria; it is rather glutinous and thus difficult to remove by mechanical means.

A third method has proven to be quite effective and simple. At periodic intervals, the contents of the microcosms are poured or siphoned into clean containers. In this way surface growth never gets a chance to build up. Because the growth is exponential in its early stages, the total amount of biomass "thrown away" by this procedure can be made to be quite small simply by carrying out the transfer procedure at sufficiently frequent intervals. We have found that weekly transfer is sufficient in large (>15 liters) systems, while more frequent transfer is probably necessary in smaller ones. This solution to the surface-growth problem may, however, generate other problems. For example, the periodic agitation of the water may disrupt nutrient cycles and thus reduce the realism of the systems, or it may reduce their replicability. Investigation of these and other possible unwanted side-effects of the transfer strategy is not in progress.

It is likely that the surface-growth problem in closed aquatic microcosms is solved. Whether this same transfer strategy will also have the effect of reducing the problem of drift of toxicants to the surfaces remains to be seen. If the toxicant drifts to the surface for biological reasons - that is, it accumulates in or on attached organisms and sticky residues - then regular transfer very likely will reduce the problem of toxicant loss to the surfaces. On the other hand, if the toxicant is physically attracted to the surface material in such a way that the accumulation is linear in time, then the effect of the pouring process on the rate of loss of toxicant will be small. Toxicants that leave the system through the air-water interface may actually exit faster as a result of the transfer process.

### Miscellaneous

Another degree of freedom in the operation of aquatic microcosms is the amount of agitation and aeration of the water. In marine microcosms, the intensity of water agitation has been shown to be an important parameter affecting the realism of the behavior of artificially confined systems, and a simple mechanical device to create a reasonably realistic water-mixing pattern has been developed (Perez et. al., 1977; Pilson et. al., 1979). The amount of turbulence in these marine microcosms is matched approximately to that in the parent estuary, with turbulence measured by the rate of dissolution of gypsum. The influence of the water agitation rate on the realism of microcosm behavior has not been studied in lake-like microcosms.

The timing, spectrum, and intensity of the illumination on a microcosm must also be selected. A 12h:12h light;dark cycle is probably adequate for most purposes and is far simpler to arrange than a realistic annual progression of diel light variation. No distorting effects resulting from the use of inexpensive high-output fluorescent lamps have been observed by us or, to our knowledge, by others. For reasons that are not completely understood, light intensities considerably lower than those outdoors (by an order of magnitude) result in reasonably realistic algal production rates and maximum biomass densities in the laboratory, under nutrient conditions that match those in natural lakes\* (Jassby et. al., 1977a; Perez et. al., 1977).

In most investigations to date the temperature of laboratory microcosms is determined by holding the air temperature in the room housing the microcosms at some constant value. At added expense, the temperature can be varied to match outdoor conditions (Perez et. al., 1977). The extent of the increased realism of microcosm behavior under such conditions has not been researched. Arguments against attempting to create a thermocline in shallow laboratory microcosms have been presented (Jassby et. al., 1977a).

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\*Lower than natural light levels may actually be desirable in that they appear to favor diatom productivity, which is often suppressed in laboratory microcosms.

The added expense and complexity needed to create realistic light and temperature conditions is probably not warranted in ecotoxicological test systems. Indeed, the temporal variation of phytoplankton and zooplankton populations in microcosms held at constant temperature and illuminated on a 12h:12h light:dark cycle over a 6-month period has been shown to resemble closely the bimodal seasonal succession pattern of certain natural systems (Jassby et.al., 1977a). Thus, natural seasonal variations in light and temperature may not be needed to produce adequately realistic seasonal biological behavior in the laboratory.

#### CONCLUSION

A great variety of options are available for the design, initiation, and operation of lake-like microcosms. The choices selected will have a large and, in many cases, a fairly well understood influence on the realism of the results of ecotoxicological studies conducted with these microcosms. Hydraulically closed systems, initiated by the whole-water-sample method, and periodically poured into clean containers to prevent surface growth will behave most like the parent lake with respect to both chemical and biotic parameters. It is also likely that such systems will provide the most realistic information about the pathways and effects of toxic substances, although here little research has taken place. The use of benthic compartments with controlled and scaled-down water-sediment exchange offers the best hope for including a benthic community in a realistic fashion in lake-like microcosms.

The influence of the choice of design, initiation, and operation options on the replicability of microcosms is probably also large, but less well understood except in a few cases. For example, replicability is often poor when organisms are initially present in small numbers, particularly when few

species are present as in gnotobiotic systems. Excellent short term replication is possible with whole-water-sample initiation; the factors responsible for occasional poor replication after several weeks of operation are not well understood.

The various options also differ considerably in their complexity and expense. Generally, no added replicability or realism is obtained by increasing the complexity and expense of microcosms. For example, whole-water-sample initiation is considerably easier than gnotobiotic initiation, closed systems are simpler and less expensive than chemostats, and surface-growth control by periodic pouring is simpler and more effective than either control by other means or the use of mathematical models to help "filter out" distortions resulting from surface growth. An important exception is the incorporation of a benthic compartment in a microcosm. Here, the easiest procedure--simply placing lake sediments on the bottom of the microcosm container--is fraught with problems and a more complex design is warranted.

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## REFERENCES

- Beyers, R. J., 1965. The pattern of photosynthesis and respiration in laboratory microecosystems. Mem. Ist. Ital. Idrobiol. 18 suppl.: 61-74.
- Dudzik, M., J. Harte, A. Jassby, E. Lapan, D. Levy, and J. Rees, 1979. Some considerations in the design of aquatic microcosms for plankton studies, Intern J. Environmental Studies, 13: 125-130.
- Grossbard, E., 1972. Problems of assessing the effects of pollutants, in: Modern Methods in the Study of Microbial Ecology, T. Rosswall, ed., pp. 457-463. Swedish Natural Science Research Council, Uppsala.
- Harte, J., D. Levy, J. Rees, and E. Saegebarth, 1979. Making microcosms an effective assessment tool, Proceedings from the Symposium on Microcosms in Ecological Research, John Giesy ed., to be published.
- Harte, J., and D. Levy, 1979. Investigation of the responses of lake waters to detritus additions, LBL-9314, submitted for publication.
- Jassby, A., M. Dudzik, J. Rees, E. Lapan, D. Levy, and J. Harte, 1977a, Production cycles in aquatic microcosms, Environmental Protection Agency Office of Research and Development Research Report EPA-600/7-77-097, National Technical Information Service, Springfield, VA, 51 p.
- Jassby, A., J. Rees, M. Duzik, D. Levy, E. Lapan, and J. Harte, 1977b, Trophic structure modifications by planktivorous fish in aquatic microcosms, Environmental Protection Agency, Office of Research and Development Research Report EPA-600/7-77-096, National Technical Information Service, Springfield, VA, 18 p.
- Maguire, B., 1971. Community structure of protozoans and algae with particular emphasis on recently colonized bodies of water, in: The Structure and Function of Freshwater Microbial Communities, J Cairns, ed. Virg. Polytechnic Inst. and Univ., pp.121-149.
- Neill, W. E., 1975. Experimental studies of microcrustacean competition, com-

- munity composition and efficiency of resource utilization, Ecology 56:809-826.
- Perez, K. T., G. M. Morrison, N. F. Lockie, C.A. Oviatt, S. W. Nixon, B. A. Buckley, and J. F. Heltsche, 1977. The importance of physical and biotic scaling to the experimental simulation of a coastal marine ecosystem. Helgolander wiss, Meereunters 30:144-162.
- Pilson, M. E., G. A. Vargo, and C. A. Oviatt, 1979. Nutrient cycles in MERL microcosms, Proceedings from the Symposium on Microcosms in Ecological Research, John Giesy, ed., to be published.
- Porcella, D. B., 1969. Continuous-flow (chemostat) assays, in Proceedings of the Eutrophication-Biostimulation Assessment Workshop, E.J. Middlebrooks et. al., eds., formerly Pacific Northwest Water Laboratory, U.S. Dept. of the Interior-Corvallis, Oregon, pp.7-23.
- Taub, F. B., 1971. A continuous gnotobiotic (species defined) ecosystem, p. 101-120. In J. Cairns (ed.), The structure and function of freshwater microbial communities. Virginia Polytechnic Institute and State University.
- Taub, F. B., 1974. Closed ecological systems. Ann. Rev. Ecol. System. 5: 139-160.
- Veldkamp, H. and J. Kuenen, 1972. The Chemostat as a model system for ecological studies, in: Modern Methods in the Study of Microbial Ecology, T. Rosswall, ed., pp. 347-356. Swedish Natural Sciences Research Council, Uppsala.
- Whittaker, R. H., 1961. Experiments with radio-phosphorous tracer in aquarium microcosms. Ecol. Monogr. 31: 157-188.



ON THE FEASIBILITY OF USING LETHAL + SEMILETHAL FREQUENCIES AND THE DISTRIBUTION  
OF LETHALS IN DROSOPHILA MELANOGASTER NATURAL POPULATIONS AS A MONITORING  
TECHNIQUE FOR THE DETECTION OF CHEMICAL MUTAGENS IN THE ENVIRONMENT.<sup>1</sup>

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Abstract: Drosophila melanogaster is frequently used in the laboratory to assess the mutagenicity of chemical compounds. Both sex-linked and second chromosomal lethal tests have been employed and the distribution of lethals induced by various chemical mutagens compared with those induced by X-ray, gamma radiation or other means. Unfortunately genetic variants which act as lethals or semilethals in homozygous condition comprise a normal component of concealed genetic variability in natural populations of this species. Some have been found to carry high mutator genes which increase the level of lethals in the populations. Also, the level of lethals and semilethals can vary over time in a single population or with distance between populations. The distribution of naturally occurring second chromosome lethals can likewise vary over time while the distribution of lethals induced by a chemical mutagen can also vary with genetic background. Data obtained on the lethal and semilethal frequencies and the distribution of naturally occurring second chromosome lethals in the S. Amherst D. melanogaster population in the 1960s and 1970s are presented and discussed as well as data on spontaneous and induced lethals. Studies on Japanese populations of D. melanogaster are also considered. Evidence available at present indicates that neither the level of lethals and semilethals nor the distribution of lethals observed in a natural population of D. melanogaster can be used as a reliable index of the presence of a chemical mutagen in the natural environment. Various reasons for this negative conclusion are explored.

Drosophila melanogaster is genetically the best studied of all the cosmopolitan species of Drosophila. Although much of the work has been with laboratory stocks, considerable progress has been made in studies on natural populations of this species. Geographical differences have been found to exist for lethals (lethal + semilethal frequencies, Ives, 1954), isozymes (Schaffer and Johnson, 1974; Band, 1975; see also Wright, 1979), biometrical traits (David et al, 1977), levels of developmental homeostasis (Tantawy and Mallah, 1961), inversions (Stalker, 1976) and morphology (Stalker, in preparation). Parsons has made it the subject of an ecological genetics study in Australia and research on all aspects of this species comprises an extensive part of the material covered in The Genetics and Biology of Drosophila, edited by Michael Ashburner and others (Vol. I a,b,c with Novitski; Vol. II with T. Wright; Vol. III with Carson and Thompson, in preparation).

The concept of marginal versus central populations, initially applied to inversions frequencies in various D. willistoni populations in different habitats (da Cunha, Burla and Dobzhansky, 1950) was found to apply to comparative differences in lethal and semilethal frequencies between Northern and Southern populations of D. melanogaster as well as to genetic changes brought about in a population in a deteriorating environment as a consequence of a persisting climatic shift (Band, 1963, 1968). With regard to isozymes, both types of populations can have comparable levels of genetic heterogeneity, and marginal and central populations of D. melanogaster behave similarly (see Band, 1975 for references).

D. melanogaster has long been a favorite laboratory tool for assessing the effects of both radiation and chemical mutagens. Abstracts presented at the European Environmental Mutagen Society in 1978 (published 1979) demonstrate that X-chromosome mutagenicity tests are frequently employed. We can thus begin our evaluation of the feasibility of using lethals and the distribution of lethals as environmental monitors for the detection of hazardous substances by considering sex-linked lethals.

X-chromosome lethals and the problem posed by hi mutator genes in populations:

One of the early questions arising in the post World War II years concerned the distribution and incidence of lethals induced by varying levels of radiation. H. J. Muller had received the Nobel Prize in 1946 for his discovery that X-rays induced mutations. The organism he used was Drosophila melanogaster. Genetic tools available continued to favor the study of sex-linked lethals (Spencer and Stern, 1948) and some regions along the X were found to be more mutable than others.

The discovery that chemicals could act as mutagenic agents (Rapoport, 1946; Auerbach and Robson, 1947) led to questions on the incidence and distribution of lethals produced by various chemical mutagens and comparisons with X-ray data. Among the early published studies, administration of suspected chemical mutagens vary from placing it in the food as in the case of formaldehyde (Rapoport, 1946) to direct injection of the chemical mutagen into adult male D. melanogaster (Fahmy and Fahmy, 1956). The latter technique added a quantitative dimension analogous to the dose/response studies on radiation. Fahmy and Fahmy (1956) showed that the distribution of sex-linked lethals induced by different alkylating mutagens differed significantly from those induced by X-rays (Spencer and Stern, 1948) although both types produced a high proportion of lethals in region 1 of the X. Up to this point we have only been talking about the distribution of lethals produced by radiation or chemical mutagens along the X chromosome of laboratory stocks of D. melanogaster.

Ives (1950) found a gene in a Florida population of this species which increased the lethal mutation rate. Other populations of D. melanogaster were also found to have such genes, including one in Michigan. He later compared the distribution of lethals on the X which were induced by the high mutator gene with the distribution of lethals induced by gamma radiation and found that mutator-induced lethals were more frequent in the ct-ras region than radiation lethals (Ives, 1959).

Nafei and Auerbach (1964) subsequently compared the distribution of sex-linked lethals induced by formaldehyde with those obtained by X-ray (Spencer and Stern, 1948), gamma rays (Ives, 1959) and a variety of other chemicals and found significant differences. They were using an Oregon-K background. They also studied the distribution of formaldehyde-induced lethals along the 2nd chromosome and found a significant deficiency of lethals in the mid-region. However, as found by Ives and Simmons (1977) a chemical mutagen can have a different effect in a different genetic background (see Table 5). Nevertheless Nafei and Auerbach (1964) also call attention to another important fact that a chemical mutagen can also be highly specific in its actions, for formaldehyde affects only the male larvae and does not induce mutations in either the female D. melanogaster larvae or in adults.

A significant excess of females to males collected would be a good indicator of the existence of sex-linked lethals in a D. melanogaster natural population. However, since hi mutator genes have been found in both Northern and Southern populations of this species, aberrant female-male ratios are not per se evidence for the existence of environmental mutagens. Furthermore, hi mutator genes, which are generally not on the X-chromosome, probably exert their influence throughout the genome. The fact that they have been found in populations as diversely distributed as Michigan and Florida complicates the interpretation that can be made on any first time sampling which reveals significant differences between lethal and semilethal frequencies from even two D. melanogaster populations from the same state or geographic region.

Second chromosome lethals and the dual problems posed by changes in both frequencies and distribution over time: The distribution of lethals along the second chromosome has been studied for spontaneous lethals (Watanabe and Oshima, 1966; Ives and Simmons, 1977), for natural populations (Paik, 1960; Watanabe

and Oshima, 1966; Ives and Simmons, 1977; Ives, this report), for mixed radiation and natural population lethals (Seto, 1963) and for chemically induced lethals (Nafei and Auerbach, 1964; Ives and Simmons, 1977).

However, the capacity for lethal and for lethal and semilethal frequencies to vary with time and distance (Ives, 1954, 1970, this report; Watanabe and Oshima, 1966; Minamori et al, 1973) as well as in distribution along the chromosome (Ives and Simmons, 1977; Ives, this report) may complicate the relative effectiveness with which autosomal lethals and their distribution can act as indicators of the presence of chemical mutagens in the environment. Even the increase observed in the frequency of lethals and semilethals in a Japanese population of D. melanogaster in the 2nd half of the 1960s (Minamori et al, 1973) may have resulted from a climatic shift affecting Japan (see Landsberg, 1971) just as changes in the S. Amherst D. melanogaster natural population during the 1960s also seem to have been associated with climatic shifts affecting the area (Ives, 1970; Band, 1969, 1972a,b; Band and Ives, 1968).

The data accumulated on the S. Amherst D. melanogaster natural population are extensive since studies were begun in 1938 (Ives, 1945). Of special interest are the studies on the distribution of lethals carried out principally in the 1970s and on lethal and semilethal frequencies and allelism rates in a variety of populations of D. melanogaster in the New York-New England area.

#### Consideration of the 1970s data:

The populations sampled: During the 1970s collections continued to be made at the traditional collecting site, Ives' porch, in September when the D. melanogaster natural population reaches its peak size and at the Markert apple storage facility in June and October or November. The apple dump associated with the storage facility appears to have been an overwintering site for at least one subpopulation of the D. melanogaster in the area and a major contributor to the peak Fall

population (Ives, 1970) during its existence prior to its termination in 1978. Additionally, a collection in Rome, N. Y. was made in 1971. In 1977 and 1978 collections were also made at Hockanum, Mass., 3 miles west of the principle collecting site and at Worthington, 10 miles west and 1100 feet up.

Methods of analyses: The usual method of analysis was employed to score the viability of 2nd chromosomes in homozygous condition. Briefly, a wild-caught male was crossed to Cy/B1 females and one F<sub>1</sub> Cy male again crossed to Cy/B1 females. The F<sub>2</sub> Cy progeny within a chromosome line are then intercrossed to yield 67% Cy and 33% +/+ if the chromosome is non-lethal in homozygous condition, 0% +/+ if the chromosome carries a lethal, less than 17% +/+ if the chromosome is semilethal in homozygous condition. Semilethals are sometimes retested if there is doubt about their viability classification. To determine the frequency of identical lethals among the lethals scored, lethal lines are crossed in all possible combinations. Lethal localization studies were carried out as described in Ives and Simmons (1977) for induced EMS lethals and tabulated in the left (0.0-49.9), middle (50.0-69.9) or right arm (70.0-108+) of the 2nd chromosome.

Additionally, the distribution of EMS induced lethals were studied in both Oregon-R and S. Amherst D. melanogaster backgrounds. For the latter 9 inversion-free non-lethal chromosome lines from the 1973 Markert collection were used. Mr. P. A. Simmons analyzed the distribution of the EMS-Markert lethals for his honors thesis work in biology.

The weather data available in the Amherst area: One of the most unique features of the Amherst area is the existence of weather data from 1838 when Prof. Snell of Amherst College began keeping records. They were continued after his death by his daughter. In the 1890s a weather station was established at the Mass. Ag. College in Amherst (now the University of Massachusetts) but was moved in the 1960s to a new location which proved less satisfactory so was discontinued. In 1948 Amherst College also established a weather station in connection with a

being offered in biology. Dr. P. T. Ives became the weather observer, having begun his own weather records on the hypothesis that climate was one of the factors affecting the D. melanogaster population in the area. Lethal and semilethal frequencies in the population remained high 1938-1946 and then declined.

In the 1970s Ives converted the Snell data to current usable figures and prepared the chapter, The Changing Climate of Amherst, in Essays in Amherst History (1976). Data on temperature means and precipitation per season for the decades, 1839-1878, are included in Table 8 merely to confirm that climate was colder and wetter in the 19th century (Landsberg, 1971) and to point out that local or regional trends may sometimes be part of a global shift.

The South Amherst population in the 1970s: Data for lethal and semilethal frequencies, and rates of allelism among lethals are given in Table 1 for the peak Fall population and in Table 2 for the Markert collections in June and October or November. In contrast to the dramatic decline in le + sle frequencies to under 20% throughout the summer of 1966 and the subsequent increase back to 35% by 1970 (see Table 2 and Fig. 1) there has been no dramatic changes in le + sle frequencies in the 1970s. As might be expected since the population begins from successful overwinterers in May or June and increases in size during the breeding season, lethal allelism rates are higher in the June population than in the Sept Fall population or at the Markert site. Reflecting the fact that a number of subpopulations contribute to the large Fall population, lethal allelism rate is generally lower in the peak Fall population (Table 1).

Although le + sle frequencies average around 35-36% in the 1970s, there have been genetic changes during this time. This is evident from the shift in the distribution of lethals along chromosome 2. From the data given in Table 3 a deficiency of lethals in the right arm and an excess of lethals in the left arm is especially evident in 1970 and 1971 in the Markert population. As found in

Table 4 the same was true for the *D. melanogaster* population from Rome, N. Y. that year. All Fall populations had the same level of le + sle frequencies, 41-42%.

If the percentages are converted to numbers for the 2 Markert lethal distributions and the Rome one and totaled, 141 lethals are found to be distributed as 80 on the left, 46 in the middle and only 15 on the right. There are obviously significantly more lethals on the left, significantly fewer on the right.

If the percentages are converted to numbers for Markert lethal distributions, 1972-1976, and totaled, we then find that 317 lethals were distributed as 116 on the left, 115 in the middle and 86 on the right. Although there is still a significant deficiency on the right ( $P < 0.025$ ) the numbers of lethals in the left arm and in the middle are more comparable than in 1970 and 1971.

Table 5 indicates that spontaneous lethals tend to occur more frequently on the right! However the distribution of chemically induced lethals shows no particular pattern. In fact, as found by Ives and Simmons (1977) EMS induced lethals show different distributions in different genetic backgrounds which may even mimic observed distributions of natural population lethals.

Table 6 shows that lethal and semilethal frequencies in *D. melanogaster* populations in the same state may vary with location. Worthington with the lowest le + sle frequency in September 1977 has an elevation of 1100 feet. However the distribution of lethals in the Hockanum sample in 1978 and the Porch sample in 1979 (Table 6) show no evidence of continuing the considerably reduced numbers of lethals in the right arm as found in the Markert distribution of lethals in 1977 (Table 2).

The South Amherst *D. melanogaster* natural population was not the only natural population of this species to have undergone genetic changes during the 1960s. Watanabe and Oshima (1966) observed a significant decline in semilethals in one Japanese population between 1964 and 1965. Minamori et al (1973) observed a decline followed by an increase in both lethals and semilethals in the Hiroshima

population. The changes observed in le + sle frequency in the peak Fall S. Amherst D. melanogaster population is plotted in relation to the observed changes in the Hiroshima population, 1961-1971, as shown in Fig. 1. Since both the warming trend peaking in Japan in the 1960s (Landsberg, 1971) and air currents blowing from the pool of warm water in the Pacific (Namais, 1969) could have contributed to warmer winter weather as well as human activities (Minamori et al, 1973), climate cannot be discounted as a factor contributing to the observed changes.

The North Pacific anomaly was one of the factors contributing to the Northeast drought. The cessation of the drought not only restored rainfall to more normal summer levels in the Amherst area the number of days in which the difference between maximum and minimum temperatures equaled or exceeded  $26^{\circ}$  F also declined (Band, 1972a,b). Both of these climatic variables had been known to be significantly related to le + sle frequency in the population (Band and Ives, 1961, 1968; Band, 1975).

Table 7 compares the average number of days in the different temperature range categories per summer, rainfall level and mean temperature for the 3 periods of population changes since 1962. In each interval the number of days in the wide range category ( $\geq 26^{\circ}$  between daily minimax temperatures) declines although rainfall and summer mean temperatures remain comparable 1967-1971 and 1972-1978.

Except for the fact that natural selection appears to act strongest against lethals on the right arm of chromosome 2, there appears to be no apparent reason why a shift to more equable temperature range conditions should also coincide with a shift in the distribution of lethals from the left toward the middle and the right.

Table 8 reinforces Table 7 in a different way; the interval during which the distribution of lethals along chromosome 2 has been studied in the Amherst

population is quite different from the interval which preceded it, 1959-1968.

Thus, if natural selection under relatively milder conditions still acts against lethals on the right and if spontaneously occurring lethals accumulate on the right, one might a priori expect chemical mutagens also to be more frequent on the right since they too would not have been acted on by natural selection. However, in the observed instances where the distribution of lethals induced by 2 different chemical mutagens in different genetic backgrounds has been studied for chromosome 2, this does not appear to be the case.

Therefore the present evidence suggests that neither the determination of lethal frequency in a natural population of D. melanogaster nor the more time-consuming process of lethal localization will reliably indicate the presence of chemical mutagens in the environment. One difficulty arises from the fact that chromosomes carrying multiple lethals do not alter the frequency of lethal-bearing chromosomes in the population. Another difficulty arises from the fact that regions defined appear to be too broad.

Only one study of the distribution of spontaneous and naturally occurring 2nd chromosome lethals has attempted to locate the specific lethal to a specific map unit comparable to the study of formaldehyde-induced lethals by Nafei and Auerbach (1964). This is the study of Watanabe and Oshima (1966) but the same localization procedures as Ives and Simmons (1977) were used. In the Japanese study however over 2000 F<sub>2</sub> cross-over and noncross-over flies were counted per determination. Considered on the basis of % left, middle or right as given in Table 5, we return to the problem that the distribution of 2nd chromosome lethals in the South Amherst D. melanogaster natural population during 1970-1979 has sometimes been similar to both the EMS-induced and the natural population lethals observed in Japanese D. melanogaster natural populations.

Hence, on the basis of the available evidence both the distribution of 2nd chromosome lethals induced by chemical mutagens and the distribution of 2nd chromosome

lethals in natural populations of D. melanogaster can vary too much to make this a reliable technique for ascertaining the presence of chemical mutagens in the natural environment.

## LITERATURE CITED

- Ashburner, M and E. Novitski, eds. 1976. The Genetics and Biology of Drosophila. Vol. 1a,b,c. Academic Press, London.
- and T. F. R. Wright. 1978. The Genetics and Biology of Drosophila. Vol. 2a, b, c. Academic Press, London.
- Auerbach, C. and J. M. Robson. 1947. The production of mutations by chemical substances. Proc. Royal Society. Edinburgh. B 62: 271
- Band, H. T. 1963. Genetic structure of populations. II. Viabilities and variances of heterozygotes in constant and fluctuating environments. Evolution 17: 307-319.
- , 1964. Genetic structure of populations. III. Natural selection and concealed genetic variability in a natural population of Drosophila melanogaster. Evolution 18: 384-404.
- , 1969. Genetic load, genetic homeostasis and balancing natural selection in a natural population of Drosophila melanogaster. Japan. J. Genet. 44 (Suppl. 1): 200-208.
- , 1972a. Minor climatic shifts and genetic changes in a natural population of Drosophila melanogaster. Amer. Nat. 106: 102-115.
- , 1972b. Further evidence of genetic and increased developmental homeostasis in a Drosophila melanogaster natural population during a minor climatic shift. Evolution 26: 116-129.
- and P. T. Ives. 1961. Correlated changes in environment and lethal frequency in a natural population of Drosophila melanogaster. Proc. Nat. Acad. Sci. 47: 180-185.
- and ———. 1968. Genetic structure of populations. IV. Summer environmental

- variables and lethal and semilethal frequencies in a natural population of Drosophila melanogaster. *Evolution* 22: 633-641.
- da Cunha, A. D., H. Burla and Th. Dobzhansky. 1950. Adaptive chromosomal polymorphism in Drosophila willistoni. *Evolution* 4: 212-235.
- David, J., C. Bocquet and M. De Scheemaeker-Louis. 1977. Genetic latitudinal adaptation of Drosophila melanogaster: new discriminative biometrical traits between European and equatorial African population. *Genet. Res.* 30: 247-255.
- European Environmental Mutagen Society, Abstracts of papers presented at the 8th annual meeting, July 11-13, 1978, Dublin. 1979. *Mutation Research* 64: 95-152.
- Fahmy, O. G. and M. J. Fahmy, 1956. Cytogenetic analysis of the action of carcinogens and tumor inhibitors in Drosophila melanogaster. V. Differential genetic response to the alkylating mutagens and X-radiation. *J. Genet.* 54: 146-164.
- Ives, P. T. 1945. Genetic structure of American populations of Drosophila melanogaster. *Genetics* 30: 167-196.
- , 1950. The importance of mutation rate genes in evolution. *Evolution* 4: 236-252.
- , 1954. Genetic changes in American populations of Drosophila melanogaster. *Proc. Nat. Acad. Sci.* 40: 87-92.
- , 1959. Chromosomal distribution of mutator- and radiation-induced lethals in D. melanogaster. *Evolution* 13: 526-531.
- , 1970. Further genetic studies of the South Amherst population of Drosophila melanogaster. *Evolution* 24: 507-518.

- Ives, P. T. 1976. The changing climate of Amherst, in Essays in Amherst History. Vista Trust, Amherst, Mass.
- and P. A. Simmons, 1977. Distribution of lethal loci on the second chromosome of South Amherst D. melanogaster. D. I. S. 52: 39.
- Landsberg, H. E. 1971. Man-made climatic changes (1971 revision). Tech. Note BN-705. Inst. Fluid Dynamics and Appl. Math. Univ. of Maryland, College Park.
- Minamori, S., K. Ito, A. Nakamura, Y. Ando and H. Shiomi. 1973. Increasing trend in frequencies of lethal and semilethal chromosomes in a natural population of Drosophila melanogaster. Japan. J. Genet. 48: 41-51.
- Nafei, H. and C. Auerbach. 1964. Mutagenesis by formaldehyde food in relation to DNA replication in Drosophila melanogaster. Z. Vererbungs1. 95: 351-367.
- Namias, J. 1969. Seasonal interactions between the North Pacific Ocean and the atmosphere in the 1960's. Month. Weather Rev. 97: 173-192.
- Paik, Y. K. 1960. Genetic variability in Korean populations of Drosophila melanogaster. Evolution 14: 329-303.
- Rapoport, J. A. 1946. Carbonyl compounds and the chemical mechanism of mutations. C. R. Acad. Sci. U.R.S.S., N.S. 54: 65 (Russian with English summary; cited by Nafei and Auerbach, 1964)
- Schaeffer, H. E. and F. M. Johnson. 1974. Isozyme allelic frequencies related to selection and gene-flow hypotheses. Genetics 77: 163-168.
- Spencer, W. F. and C. Stern, 1948. Experiments to test the validity of the linear R-dose/mutation frequency relation in Drosophila at low dosage. Genetics 33: 43-74.
- Seto, F. 1963. Distribution of recessive lethals on the second chromosome D. I. S. 37: 127-128.

- Stalker, H. D. 1976. Chromosome studies in wild populations of D. melanogaster.  
Genetics 82: 323-347.
- Tantawy, A. C. and G. S. Mallah, 1961. Studies on natural populations of  
Drosophila. I. Evolution 15: 1-14.
- Watanabe, T. K. and C. Oshima. 1966. Distribution of natural lethal genes  
on the second chromosome of Drosophila melanogaster. Japan. J. Genet.  
41: 367-378.
- Wright, S. 1978. Evolution and the Genetics of Populations. Vol. IV. Variability  
within and among Natural Populations. Univ. of Chicago Press, Chicago.

Table 1. Lethal + semilethal frequencies and allelism rates among lethals from second chromosomes from the Fall population of *D. melanogaster* in Amherst, Mass., 1938-1979. The number of collections are also indicated where pooled data are given from previous publications on the population.

Year	# col- lections	# chromo- somes	le + sle	%	Cross- tests	allelic	%
1938-46	4	528	258	48.8	2307	9	0.39
1947-53	7	1458	514	35.2	6584	59	0.90
1954-61	4	895	279	31.2	2251	15	0.67
1962-66	5	637	177	27.8	469	2	0.43
1967-71	5	1512	523	34.6	4243	210	2.27
1972		271	96	35.5	1430	53	3.71
1973		243	86	35.4	1711	7	0.41
1974		285	95	33.3	2078	20	0.96
1975		271	92	34.0	1596	6	0.38
1976		241	91	37.8	2075	55	2.65
1977		188	75	39.9	1377	4	0.29
1978		160	72	45.0	1225	9	0.73
1979		309	106	34.3	1225	12	0.98

Collections from 1938 through 1971 have been grouped according to intervals of suspected genetic changes in the population, for which see Ives (1954, 1970), Band (1964, 1972a,b), Band and Ives (1968).

Table 2. Lethal + semilethal frequencies and allelism rates among lethals from second chromosomes from the D. melanogaster natural population at the Markert Apple Storage site. Part of the data, 1966-69, has appeared in Ives (1970, Table 2).

Year	Date	# chromo- somes	le + sle	%	Cross- tests	alleles	%
1966	June 6	369	63	17.1	1378	67	4.9
	Nov. 14	210	41	19.5	777	5	0.6
1967	June 16	566	129	22.8	1035	211	20.4
	June 29	322	81	25.2	1326	131	9.9
	Nov. 7	208	57	27.4	1223	11	0.9
1968	June 21	263	73	27.8	1378	470	34.1
	July 31	246	73	29.7	1769	538	30.5
	Oct. 31	246	77	31.3	2415	97	4.0
1969	June 20	302	97	32.1	3160	509	16.1
	Oct. 29	223	75	33.6	1486	5	0.3
1970	June	304	116	38.2	1326	141	10.6
	Nov.	265	96	36.2	3231	66	2.0
1971	June	376	133	35.4	7123	308	4.3
	Nov.	255	106	41.6	3651	99	2.7
1972	June	523	174	33.3	6427	375	5.83
	Oct.	373	131	35.1	1770	30	1.7
1973	June	312	157	50.3	1375	109	7.7
	Oct.	340	130	38.3	1427	9	0.63
1974	July	273	69	25.4	1768	41	2.3
	Nov.	289	91	31.7	2278	24	1.0
1975	Nov.	226	92	34.6	1595	10	0.6
1976	Oct.	252	88	34.9	1652	23	1.4
1977	Oct.	255	92	35.9	1644	41	2.5

Table 3. Distribution of lethals along chromosome 2, Markert D. melanogaster population, 1970-1977

Year	Month	# lethals	% Left (0 - 49.9)	% Middle (50.0-69.9)	% Right (70.0 - 108.0)
1970	Oct.	41	51.2	29.2	19.5
	Nov.	50	24.0	58.0	18.0
1971	June	46	50.0	39.1	10.9
	Nov.	50	64.0	28.0	8.0
1972	June	40	45.0	30.0	25.0
	Oct.	73	39.8	30.1	30.1
1974	Nov.	93	34.4	40.9	24.7
1975	Nov.	70	28.6	41.4	30.0
1976	Oct.	41	41.3	34.8	24.9
1977	Oct.	44	43.2	40.9	15.9
Salivary chromosome bands		2000	30 ±	40 ±	30 ±

Table 4. Lethal + semilethal frequencies, allelism frequencies among lethals and distribution of lethals along chromosome 2 for D. melanogaster populations from Amherst, Mass. and Rome, N. Y. in 1971.

Source	Month	# chromo- somes	# le + sle	%	Cross tests	alleles	%
Markert	June	376	132	35.4	7123	308	4.3
	Nov.	255	107	41.6	3651	99	2.7
Porch	Sept.	272	112	41.2	2078	58	1.2
Rome, N.Y.	August	184	77	41.8	1273	22	1.7

Source	Month	Distribution			
		# lethals	% Left	% Middle	% Right
Markert	June	46	50.0	39.1	10.9
	Nov.	50	64.0	28.0	8.0
Rome	Aug.	45	55.6	31.1	13.3

Table 5. Distribution of spontaneous, induced and other natural population lethals including later collections from Massachusetts *D. melanogaster* populations

Source	# lethals	% Left (0.0-49.9)	% Middle (50.0-69.9)	% Right (70.0-108+)
<b>A. Spontaneous</b>				
Oregon-R	57	28.1	28.1	43.8
Amherst	33	27.3	33.3	42.4
Japan*	26	30.7	26.9	42.3
<b>B. Induced, EMS**</b>				
Oregon-R	80	33.8	31.2	35.0
1973 Markert	81	42.0	42.0	16.0
<b>C. Induced, Formaldehyde***</b>				
Oregon-K	126	41.3	23.8	34.9
<b>D. Natural Populations</b>				
1964 Japan*	73	31.5	45.2	23.3
1978 Hockanum, Mass.	74	39.2	37.8	23.0
1979 Amherst (Porch)	61	42.6	32.8	24.6

\* data from Watanabe and Oshima (1966)

\*\* data from Ives and Simmons (1977). The difference in the distribution of the two sets of EMS induced lethals is significant at the 2% level.

\*\*\* data from Nafei and Auerbach (1964). Based on an expectation of equal numbers in all 3 regions, the distribution is significant at the 0.025 level.

Table 6. Comparison of lethal + semilethal frequencies and lethal allelic rates for 2nd chromosome lethals from Fall 1977 Massachusetts D. melanogaster natural populations and Fall 1978 populations.

Site	Month	# chromo- somes	#le + sle	%	Cross tests	alleles	%
A. 1977							
Worthington	Sept.	322	94	29.2	1326	9	0.68
Hockanum	Sept.	301	98	32.6	1128	10	0.89
Porch	Sept.	188	75	39.9	1377	4	0.29
Markert	Oct.	255	92	35.9	1644	41	2.49
B. 1978							
Hockanum		287	107	37.3	1225	5	0.41
Porch		160	72	45.0	1225	9	0.73

Hockanum is about 5 km west of the principle collecting site (Porch), Markert about 5 km east of it. Worthington is 20 miles west and 1100 feet up.

Table 7. Number of days per summer having a narrow ( $\leq 20^{\circ}$  F), intermediate ( $21^{\circ} - 25^{\circ}$ F) or wide ( $\geq 26^{\circ}$  F) range between daily minimax temperatures in each interval of suspected genetic changes in the S. Amherst D. melanogaster natural population. Average total rainfall and mean temperature per summer are also given.

Category	1938- 1946	1947- 1953	1954- 1961	1962- 1966	1967- 1971	1972- 1978
no.	9	7	8	5	5	7
Narrow	30.8	24.3	28.3	19.2	24.4	33.0
Inter- mediate	26.3	24	26	20.6	24.2	26.1
Wide	34.9	43.7	37.7	52.2*	43.4	32.9
Rainfall (in inches)	12.66	9.71	12.71	8.59	12.24	12.41
mean $t^{\circ}$ (F)	69.1	70.3	69.7	69.0	70.3	69.8

\*  $P < 0.05$

Table 8. Annual and Seasonal Temperatures and Precipitation by Decade in Amherst, Mass.: 1839-1878 compared with 1939-1978.

Decade	Annual	Winter	Spring	Summer	Fall
A. Temperature in ° Fahrenheit					
1839-1848	46.4	24.3	44.9	67.8	48.4
1849-1858	45.3	22.8	42.5	67.3	48.4
1859-1868	45.6	23.2	43.7	67.0	48.7
1869-1878	46.2	24.2	43.7	68.4	48.4
mean	45.9	23.6	43.7	67.6	48.5
1939-1948	48.0	24.9	46.4	69.2	51.5
1949-1958	49.1	28.4	46.9	69.6	51.5
1959-1968	47.6	24.5	46.0	68.7	51.2
1969-1978	48.3	25.2	47.3	70.3	51.3
mean	48.2	25.8	46.7	69.4	51.4
B. Precipitation in Inches					
1839-1848	42.41	9.93	9.71	11.82	10.69
1849-1858	45.39	10.01	11.15	12.62	11.86
1859-1868	46.19	9.71	11.59	13.92	11.25
1869-1878	45.23	8.96	10.74	13.27	11.80
mean	44.81	9.65	10.76	12.91	11.40
1939-1948	41.13	8.46	11.48	11.58	9.42
1949-1958	42.93	9.95	11.58	10.74	10.79
1959-1968	38.00	8.34	8.81	10.85	9.63
1969-1978	46.61	11.75	11.41	12.33	11.12
mean	42.17	9.63	10.82	11.38	10.24

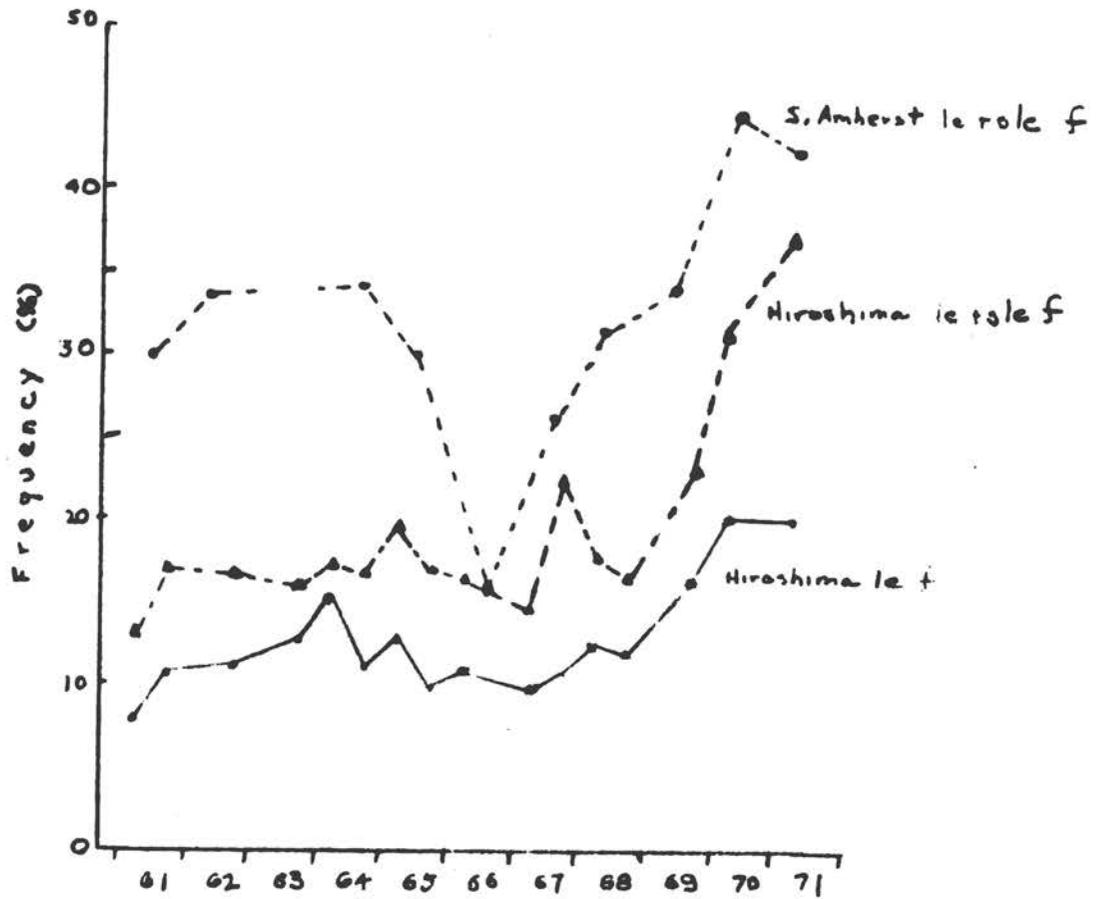


Fig. 1. Comparison of the frequencies of lethals and semilethals in the peak Fall population of *D. melanogaster* in S. Amherst, Mass (U.S.A.) with those of the Hiroshima population of this species during 1961-1971. Data on the Japanese population from Minamori et al (1973).



BACKGROUND PAPER

CHEMICALS AND WETLANDS

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Abstract: This paper addresses the types of information needed to do a comprehensive environmental assessment of wetland ecosystems, with emphasis on characteristics which are important in determining the fate of toxic chemicals. The major types of chemicals are categorized, and their probable influences are discussed.

## Background

The physical features of a wetland ecosystem can be categorized into the areas of topography, hydrology, soil structure, and spatial patterns. In addition to these macroscopic compartments, one can add the physical microscopic compartments of the dissolved nutrient status of the wetland waters and their interaction with the soil chemistry. The complex interactions among all of these constituent parts of the wetland ecosystem determine the character and classification of a particular wetland. The topography of a wetland ecosystem is frequently very flat, with sphagnum bogs being a notable exception. This flatness leads to the necessary water retention which characterizes all wetlands. The water budget is the key feature to which all physical wetland functions can be tied. The opposing processes of precipitation and evapotranspiration are the interactions with atmospheric water. Beneath its surface, the wetland may also be exchanging water with subterranean aquifers - recharge or discharge. These interchange processes affect the stored water pool and its water chemistry. They also have a direct effect on the nature of the biota in a particular wetland.

Wetlands vary widely in biological characteristics. In northern temperate regions, certain wetlands are dominated by a complete cover of mosses, with only a few scattered shrubs; or may be dominated by sedges. Other habitats may

have a cover of cattail, or bulrush. Forested wetlands in the southern areas of the U.S. contain deep water swamps. Most wetland ecosystems support a prolific growth of plants.

The vegetation of wetland ecosystems supports two types of food webs. First, there are organisms which graze directly on live plant tissues. These organisms include certain mammals, birds, amphibians, fish, and invertebrates. The second web is composed of organisms which utilize detritus or the bacteria or fungi which colonize the surface of particulate organic matter. The combined effect of these two food webs is that wetlands have a large diversity and high density of organisms. The discharge of pollutants affects the wetland biota to some extent, so any such discharge must be carefully evaluated.

#### Assessment Goals

A natural wetland receives inputs of water, dissolved nutrients, suspended solids, and organisms. The same wetland generally discharges the same materials in altered quantities, at perhaps different points in time, to its surroundings. The wetland functions so as to retain its generally wet character, and the attendant biotic and abiotic processes and constituents. Discharge of a pollutant into a wetland ecosystem will alter, to some extent, its function, characteristics, and output. This fact gives rise to two types of questions concerning a particular stress on the wetland:

ecological questions relating to the character and function of the wetland, and water renovation questions relating to the quality of the water discharged from the wetland.

The water renovation question is perhaps best addressed from the so called "black box" approach, which considers inputs, outputs, and accumulations within the wetland. Detailed knowledge can be gained only by considering localized events within the wetland pertaining to nutrient cycling, biotic and abiotic processes, and the associated environmental controls. An ecosystem experiences both regular periodic and random variability. One must consider daily, seasonal, climatic, and geological time scales. The consideration of time alone is not sufficient to understand the ecosystem since questions of spatial patterns within the wetland are also of great importance. These may pertain to distance effects related to water movement, or to species composition, or to depth into the underlying strata. Knowledge gained in any of these areas must be capable of extrapolation to other ecosystem types and with respect to time. This requires that gathered information be organized in such a way that computations can be made of potential effects in new situations and at different points in time.

Any attempt to ascertain the impact of a chemical on the wetland, or to trace its progress in the wetland, must be preceded by an understanding of the current status and function of that ecosystem. These may in turn be categorized:

- 1) hydrological patterns; 2) water quality; 3) soil type;
- 4) chemical status of soil, water, flora and fauna; 5) types and relative abundance of algae, invertebrates, and bacteria;
- 6) types and geographical distribution of vegetation;
- 7) vertebrate use patterns; 8) human use patterns; and
- 9) historical features.

Water movement may be quantified by measurements of evapotranspiration, precipitation, streamflows, and subsurface water movements. These should be complemented with water level records for the wetland and adjacent water bodies; and a surface elevation study aids in interpreting measurements of surface flow patterns within the wetland. This water budget data is a vital framework for interpreting measures of water-borne contaminants. Concentrations of various materials may then be converted to budgets which detail inputs, outputs and accumulations. Water quality, in terms of nutrients, suspended solids, toxic chemicals and other dissolved materials, then becomes meaningful.

Soils are an essential part of the wetland ecosystem, both as a locus for microbial processes and as a reservoir of contained and adsorbed chemicals. There is no doubt that a wide variety of soils adsorb trace constituents at rapid rates, but with relatively small capacity. The types and loading status of the soil compartment are thus important. Similarly, the flora and fauna may contain elevated or depressed levels of the compound in question, and should be

assayed. The mobile organisms, such as algae and invertebrates can be especially important in the movement of trace compounds. Vegetation may vary geographically in response to chemical additions: cattails may invade in response to chemical stress, for example.

The ultimate fate of a particular chemical may be in the upper trophic levels, either as a vector or a sink for that substance. Humans rarely use wetlands, but, along with other large vertebrates, may modify the ecosystem. Finally, a historical perspective is needed. Some wetlands may be "loaded" with a particular chemical because of past management practices.

#### Wetland Features Relevant to Chemical Interactions

Wetlands appear to be strongly buffered against the stress of chemical additions. Dissolved materials are effectively absorbed in many cases, and suspended solids are exchanged. The added chemicals have an impact on the ecosystem which varies from site to site. The plant and animal communities undergo changes which range from barely detectable to dramatic.

There is a commonality among all studies which display poor to mediocre chemical uptake. This commonality is deep, fast-moving waters, channelized situations, and poor contact with an organic soil substrate. Thus it would appear that

thorough contact with a soil substrate is a requirement for initial chemical immobilization and particulate filtration. Systems such as those reported by Stanlick,<sup>9</sup> Small and Wurm,<sup>10</sup> and Kadlec et al.,<sup>11</sup> appear to have met this constraint. Odum et al.<sup>12-14</sup> also appear to have met this constraint by downward flow of the wastewater through the organic mat in the cypress domes. This is not unreasonable in view of the fact that removal and filtration processes are rate processes which depend in part on sufficient time of contact with those components of the ecosystem that are performing the purification function.

A wetland intentionally hydrologically overloaded would produce a reduction in removal potential. Kadlec and Tilton<sup>15</sup> show that the degree of nutrient removal of the Bellaire wetland is a function of the total amount of nutrient added to the system as well as the hydrologic regime, so that hydrologic condition alone does not determine water treatment potential. Rather, it is the level of contamination of incoming water coupled with residence time and contact between the added wastewater and the wetland ecosystem substrates that determine the efficiency of removal.

#### NITROGEN AND PHOSPHORUS

Those chemicals which are nutrients display a cyclic behavior. The uptake of nutrients can be partitioned into four general classes as follows: 1) vascular plant uptake,

2) algal uptake, 3) bacterial and fungal uptake and transformation, and 4) sediment processes (sorption, ion exchange, precipitation, etc.). There is little quantitative information regarding the relative importance of the components to nutrient sorption in wetlands.

Although quantitative data are scarce, nutrient uptake can be described in general terms. During the growth and metabolism of vascular plants, nutrients are absorbed to supply the requirements of the plants. These nutrients may be obtained directly from the water or from the surface of exchange sites in the soil. Upon the death and decay of these plants, a certain amount of the absorbed nutrients is released depending on the particular species of plant. The portion of nutrients retained in the organic matter is deposited as peat. The released nutrients recycle through the ecosystem or are transported to adjacent ecosystems. Algal uptake operates in a very similar manner, except that the release of nutrients upon the death of algae may be more complete because of the lack of structural tissues in algae.

Microbial processes are exceedingly important in the nutrient uptake processes in wetlands. Many microorganisms absorb nutrients during growth, thus enriching the protein content of the organic matter upon which they are growing. Perhaps more significant is the transformation of inorganic forms of nitrogen within wetland ecosystems. Nitrosomonas transforms  $\text{NH}_4^+ \text{-N} \rightarrow \text{NO}_2^- \text{-N}$ , and Nitrobacter transforms

$\text{NO}_2^- \text{-N} \rightarrow \text{NO}_3^- \text{-N}$ . The process is autotrophic, requiring a well-aerated environment, suggesting that if it occurs at all in wetlands, it occurs in the relatively well-aerated surface waters. Microbial nitrate reduction may be one of the most important processes in wetlands relative to the utilization of these ecosystems for wastewater renovation.<sup>16</sup> Nitrate is denitrified ( $\text{NO}_3^- \rightarrow \text{N}_2$  and  $\text{NO}_2^-$ ) under appropriate conditions by several genera of facultative anaerobic bacteria.

The length of time of nutrient retention in wetland ecosystems is an issue clouded by contradictory results. Lee et al.<sup>17</sup> suggest that any phosphate absorbed by the wetland in the summer is released in the spring during runoff of snow-melt water. Studies of wetlands in Michigan, however, do not support this view.<sup>15,18,19</sup> There tends to be a larger amount of nutrients transported during spring runoff, but the amount is not equivalent to that absorbed by the wetland during the summer.

#### HEAVY METALS

Aquatic and semiaquatic plants absorb heavy metals, and interactions between dissolved ions and sediments cause metal accumulations in the soil as well. The pattern varies among wetlands and heavy metals, but it appears that a greater proportion of heavy metals is sedimented rather than absorbed by plants. The impact of this distribution

on the food web in wetlands and bordering lakes and streams has not been thoroughly studied. Once incorporated into plant tissues, detritus from the plants will have a high concentration of heavy metals, entering the detritus food web and thus contaminating the aquatic environment.

In wetlands, the sediments may act as a second sink for absorption of heavy metals. Of the Pb added to a salt marsh in Massachusetts, only 6-8% was absorbed by the grasses, the remainder was in the sediments.<sup>20</sup> Zn and Cd were also absorbed by live plants, but less of these elements were retained in the sediments. Surface sediments from a wetland system in Ontario, Canada showed increased concentrations of Pb, Cr, Ni, Cu, and Zn compared to deeper layers.<sup>21</sup> The metals were attributed to a sewage plant discharge into the wetland. Similarly, sediment cores from cypress domes receiving effluent in Florida show an accumulation of heavy metals in surface sediments.<sup>22</sup>

In the salt marshes, Banus et al.<sup>20</sup> found that fiddler crabs and mussels in experimental plots had higher tissue concentrations of Cd than organisms in control plots, but there was no difference among treatments for tissue concentrations of Zn or Pb. Besides these detritus-feeding organisms, there is a possibility that animals which graze directly on plant material may become contaminated. Waterfowl, mammals, and many insects graze directly on plant parts and by doing so may incorporate unknown amounts of

metals into their tissues.

#### REFRACTORY CHEMICALS

These materials may enter wetlands by direct or indirect routes. The materials are generally hydrocarbons or halogenated hydrocarbons which possess persistent, toxic properties. Because they are complex molecules, analytical procedures are often difficult. Further, the concentrations of the materials are often very low.

The effects of petroleum hydrocarbons introduced into wetlands have been the subject of some study. Analyses of marsh muds and organisms collected after a one-time spill showed some uptake of hydrocarbon material, but a general persistence of heavier materials<sup>23,24</sup> indicate that microbial processes are responsible for the dissolution of hydrocarbons from chronic petroleum input to a shallow water marsh.

Marsh plants have been determined (Seidel<sup>25</sup>) to be capable of removing a variety of organic chemicals from waters. Similar results have been reported by Wolverton and McKown<sup>26</sup> for phenol uptake. It appears that both microbial activity and uptake by higher plants can function effectively to remove at least some complex organic chemicals.

Lunz<sup>27</sup> determined that, though sediments and plants contained measurable concentrations of substances such as alpha-chlordane, there was no apparent correlation between the sediments and concentrations within plant tissues.

## OTHER WATER QUALITY PARAMETERS

In addition to concentrations of inorganic nitrogen and various forms of phosphorus, several other parameters are frequently used to evaluate water quality. These parameters include biochemical oxygen demand (BOD), chemical oxygen demand (COD), suspended solids, and methylene blue active substances, to name a few. Furthermore, because wetland ecosystems and many wetland plant species are sensitive to changes in pH, chloride, and cation concentrations, these parameters are frequently measured.

A common parameter measured in wastewater treatment facilities is BOD. Wastewater treatment facilities which utilize wetlands in some stage of the wastewater renovation have excellent BOD reductions. Average  $(BOD)_5$  in effluent entering a marsh/pond system was 170 mg/l compared to 19 mg/l leaving the system.<sup>28</sup> BOD decreased from 49.3 to 5.3 mg/l in effluent pumped into a wetland in Massachusetts;<sup>29</sup> BOD decreased from 117.7 to 2.7 mg/l after passage through a sedge meadow in Canada;<sup>30</sup> and water passing through a cattail marsh in Wisconsin showed an 80% reduction in BOD.<sup>31</sup> Similarly, various ponds planted in rushes or reeds are capable of reducing the BOD of effluent by more than 90%.<sup>32,33</sup> These values vary with retention time in the system, but it is clear that the BOD level of effluent is reduced after passing through wetland ecosystems of many different types.

COD in effluent water is similarly affected. DeJong<sup>33</sup> reported a COD reduction of 86.8% in effluent with a concentration of 530 mg/l which had passed through a pond dominated by Scirpus lacustris. In Wisconsin,<sup>31</sup> COD was reduced 43.7% from an initial concentration of 106 mg/l, and Small<sup>28</sup> reported COD was reduced from 495 to 58 mg/l. The former system was a cattail marsh; the latter a cattail marsh/shallow pond. As emphasized earlier, wetland ecosystems have natural levels of material, and these levels may sometimes be above effluent concentrations. In Michigan, Tilton et al.<sup>34</sup> reported COD concentrations of 40 mg/l in secondarily treated wastewater compared to 100 mg/l in the natural wetland surface water.

The wetland performs much like a buffer with regard to parameters, such as pH, alkalinity, and hardness. The nature of the effluent becomes adjusted to the wetland. Tilton et al.<sup>35</sup> showed pH of effluent to fall to natural levels within 10 m of a discharge site. Similar results for alkalinity and hardness have been reported elsewhere. Chloride concentrations tend to be reduced very little during flow across a wetland, primarily because of the biologically conservative nature of this element.

In a natural condition, a wetland receives particulate matter from two primary sources: runoff from surrounding upland and litterfall of particulate matter from the emergent vegetation in the wetland. The decomposition of the litter within the wetland itself can lead to fine

particulate organic matter which is ultimately capable of transport from the wetland during high runoff water conditions. Because wetland waters are stagnant or at least very slow moving, methods of sampling for suspended solids become very difficult. These materials may, however, transport major amounts of toxic chemicals.<sup>36</sup>

Little is known about sulfur compounds. Wetlands are capable of producing hydrogen sulfide and other forms of gaseous sulfur. In some cases, bacteria are involved, but little quantitative information exists about the influence of biotic or abiotic factors on the processes. Studies of the hydrogen sulfide content of a Florida cypress wetland (Odum and Ewel<sup>37</sup>) showed that hydrogen sulfide, like other water quality parameters in wetlands, varies with location and time. Many processes involving sulfur have been observed but not quantified.

#### PATHOGENS

The two "toxins" of interest are viruses and bacteria, the latter usually being measured as coliform and fecal coliform counts. Some investigators (Boyt, et al.,<sup>38</sup> for example) examined the use of fecal streptococci as a water quality indicator.

A wide variety of studies have been conducted on wetlands receiving treated waste water. For example, Yonika and Lowry<sup>29</sup> determined an increasing ratio of fecal streptococci to fecal coliforms as the water proceeded from input to output. Boyt, et al.<sup>38</sup> found just the opposite.

Odum, et al.,<sup>39</sup> found that "the soil in the area of the control and experimental [cypress] domes seems to be filtering out nearly all of the coliforms...". However, the approximate 100-fold decrease between fecal coliform numbers in the entering treated sewage and those in the standing water in the experimental dome probably resulted from dilution and mortality. Despite the reduction in fecal coliform counts, the experimental dome was contributing more fecal coliforms to ground water than the control dome not receiving sewage effluent.

A common feature of bacteriological studies appears to be extreme variability in measurements made on wetland waters (Kadlec;<sup>40</sup> Grant and Patrick;<sup>41</sup> and Spangler<sup>31</sup>). This makes it difficult to interpret transect type information, except in those instances in which there are orders of magnitude reductions or changes in the numbers of bacteria.

The survival of viruses in a wetland is important from the view of public health. The extreme difficulty and expense of conducting good virological assays have prevented extensive studies at wetland sites. The impetus for the few existing studies came from the addition of waste water to wetlands. An extensive study of cypress wetlands receiving effluent was made by Wellings.<sup>42</sup> The study found few viruses in the input water and no viruses in the wetland receiving water body.

## Conclusions

A few generalities can be concluded. Wetlands are buffered against chemical changes. They appear to maintain a stationary operating point even in the face of natural and man-made stresses. Wetland water quality varies with time (diurnal, seasonal, historical) and water flow distance. The variability makes measurements difficult, and lengthens the period of study required to successfully develop conclusions. Hydrologic regime is an important physical factor governing chemical transport. If we do not understand all components of the water budget, we cannot understand variation of chemical parameters. Sediment appears to be the action zone for most uptakes and initial storages within the wetland. Cycling determines localized and instantaneous concentrations within the wetland. Uptake by plants, incorporation into their tissues, and subsequent return to the sediments is an important route for many materials on a time scale of months. Epiphytes participate in a similar cycle, but on the time scale of hours. Tertiary waste water treatment is effective in a number of wetlands. Although these conclusions may appear to be simplistic, they were not recognized as recently as the late 1960's.

Certain generalities concerning research needs emerge. More mass balances are required in order to properly interpret wetland function; hence, there is a need for detailed information on wetland hydrology. We also need to focus on the

rate at which a particular chemical is altered in a particular wetland. More information on the ultimate sinks of the materials that arrive in a wetland by way of water is required. If a material is removed from the water, we need to know to what the material has been converted. In some cases, a substance may be degraded into biologically inactive and innocuous materials; in other cases, a noxious substance may be stored in one of the wetland compartments.

Selected References

## I. General

1. Workshop Report on Research Priorities for Wetland Ecosystem Analysis, The National Wetlands Technical Council, Report to the National Science Foundation, December 1978, 68 pp.
2. Wetland Functions and Values: The State of our Understanding, American Water Resources Association, Minneapolis, Minnesota, 1979, 674 pp.
3. Freshwater Wetlands: Ecological Processes and Management Potential, R. E. Good, D. F. Whigham and R. L. Simpson, Eds., Academic Press, New York, 1978, 378 pp.
4. Freshwater Wetlands and Sewage Effluent Disposal, Tilton, D. L., Kadlec, R. H., and Richardson, C. J., Proceedings of the NSF/RANN Conference on Freshwater Wetlands and Sewage Effluent Disposal. University of Michigan, Ann Arbor, Michigan, May 10-11, 1976, 343 pp.
5. Environmental Quality Through Wetland Utilization, M. A. Drew, Ed., Proceedings of a Symposium sponsored by the Coordinating Council on the Restoration of the Kissimmee River Valley, Tallahassee, Florida, March 1978, 243 pp.
6. Wetlands: Ecology, Values and Impacts, C. B. DeWitt and E. Soloway, Eds., Proceedings of the Waubesa Conference, Madison, Wisconsin, June 1977, 388 pp.
7. Proceedings of the Seminar on Aquaculture Systems for Wastewater Treatment, University of California, Davis, California, September 1979.

## II. Specific

8. Kadlec, R. H. and D. L. Tilton, "The Use of Wetlands as a Tertiary Treatment Procedure." Critical Reviews in Environmental Control, Vol. 9, Issue 2, 1979.
9. Stanlick, H. T., "Treatment of secondary effluent using a peat bed, in Proceedings of a National Symposium on Freshwater Wetlands and Sewage Effluent Disposal, Tilton, D. L., Kadlec, R. H., and Richardson, C. J., Eds., University of Michigan, Ann Arbor, 1976, 257.

10. Small, M. and Wurm, C., Meadow/Marsh/Pond System, Data Report, Brookhaven National Laboratory, U.S. Energy Research and Development Administration, Upton, N.Y., 1977.
11. Kadlec, R. H., Tilton, D. L., and Kadlec, J. A., Feasibility of Utilization of Wetland Ecosystems for Nutrient Removal from Secondary Municipal Wastewater Treatment Plant Effluent, Semi-Annual Report No. 5, University of Michigan, Ann Arbor, 1977.
12. Odum, H. T., Ewel, K. C., Ordway, J. W., and Johnston, M. K., Cypress Wetlands for Water Management, Recycling and Conservation, Second Annual Report, University of Florida, Gainesville, 1975.
13. Odum, H. T., Ewel, K. C., Ordway, J. W., and Johnston, M. K., Cypress Wetlands for Water Management, Recycling and Conservation, Third Annual Report, University of Florida, Gainesville, 1976.
14. Odum, H. T. and Ewel, K. C., Cypress Wetlands for Water Management, Recycling and Conservation, Fourth Annual Report, University of Florida, Gainesville, 1978.
15. Kadlec, R. H. and Tilton, D. L., Monitoring Report on the Bellaire Wastewater Treatment Facility 1977, Utilization Report No. 2., University of Michigan, Ann Arbor, 1978.
16. Engler, R. M. and Patrick, W. H., Jr., Nitrate removal from floodwater overlying flooded soils and sediments, J. Environ. Qual., 3, 409, 1974.
17. Lee, G. F., Bentley, E., and Amundson, R., Effects of marshes on water quality, in Coupling of Land and Water Systems, Hasler, A. D., Ed., Springer-Verlag, New York, 1975, 105.
18. Kadlec, R. H. and Tilton, D. L., Monitoring Report on the Bellaire Wastewater Treatment Facility 1976-77, Utilization Report No. 1, University of Michigan, Ann Arbor, 1977.
19. Kadlec, R. H. and Tilton, D. L., unpublished research results, University of Michigan, Ann Arbor, 1978.
20. Banus, M. D., Valiela, I., and Teal, J. M., Lead, zinc and cadmium budgets in experimentally enriched salt marsh ecosystems, Estuarine Coastal Mar. Sci., 3, 421, 1975.

21. Mudroch, A. and Capobianco, J. A., A Preliminary Study on a Natural Marsh Receiving Sewage Treatment Plant Effluent, Canada Centre for Inland Waters, Burlington, Ontario, 1978.
22. Carriker, N. and Brezonik, P. L., Heavy metals, in Cypress Wetlands for Water Management, Recycling and Conservation, Third Annual Report, University of Florida, Gainesville, 1975, 372.
23. Burns, K. A. and J. M. Teal, Hydrocarbon Incorporation into the Salt Marsh Ecosystem from the West Falmouth Oil Spill. Technical report, Woods Hole Oceanographic Institution, Massachusetts, 1971.
24. Whelan, T., J. T. Ishmael, and W. S. Bishop, Long-Term Chemical Effects of Petroleum in South Louisiana Wetlands. I. Organic carbon in sediments and waters. Marine Pollution Bulletin 7(8), 1976.
25. Seidel, K., Reinigung von Gewässern durch höhere Pflanzen. Naturwissenschaften. 53 Jg, Heft 12, 1966.
26. Wolverton, B. C. and M. M. McKown, Water Hyacinths for Removal of Phenols from Polluted Waters. Aquatic Botany 2, 1976.
27. Lunz, J., The Uptake of Toxic Substances by Wetland Soils and Plants. Paper presented at National Symposium on Wetlands, Lake Buena Vista, Florida, 1979.
28. Small, M. M., Marsh/Pond System, Data Report, Brookhaven National Laboratory, U.S. Energy Research and Development Administration, Upton, N.Y., 1976.
29. Yonika, D. and Lowry, D., Wetland Disposal of Wastewater Treatment Plant Effluent, IEP, Inc., Wayland, Mass., October 24, 1977 to October 24, 1978.
30. Hartland-Rowe, R. and Wright, P. B., Effects of sewage effluent on a swampland stream, Verh. Int. Ver. Limnol., 19, 1575, 1975.
31. Spangler, F. L., Sloey, W. E., and Fetter, C. W., Jr., Wastewater Treatment by Natural and Artificial Marshes, Final Report to EPA, University of Wisconsin, Oshkosh, 1976.
32. Seidel, K., Macrophytes and water purification, in Biological Control of Water Pollution, Tourbier, J. and Pierson, R. W., Jr., Eds., University of Pennsylvania Press, Philadelphia, 1976, 109.

33. de Jong, J., The purification of wastewater with the aid of rush or reed ponds, in Biological Control of Water Pollution, Tourbier, J. and Pierson, R. W., Jr., Eds., University of Pennsylvania Press, Philadelphia, 1976, 133.
34. Tilton, D. L., Recovery of nutrients from peatlands used for tertiary treatment, in Proceedings of a National Symposium on Freshwater Wetlands and Sewage Effluent Disposal, Tilton, D. L., Kadlec, R. H., and Richardson, C. J., Eds., University of Michigan, Ann Arbor, 1976, 307.
35. Tilton, D. L., Kadlec, R. H., and Linde, J., Surface water chemistry and element budgets of a wetland used for wastewater treatment, in Feasibility of Utilization of Wetland Ecosystems for Nutrient Removal from Secondary Municipal Wastewater Treatment Plant Effluent, Semi-Annual Report No. 5, Kadlec, R. H., Tilton, D. L., and Kadlec, J. A., Eds., University of Michigan, Ann Arbor, 1977.
36. Boto, K. G. and W. H. Patrick, Jr., The Role of Wetlands in the Removal of Suspended Sediments. Paper presented at National Symposium on Wetlands, Lake Buena Vista, Florida, 1979.
37. Odum, H. T. and K. C. Ewel, Cypress Wetlands for Water Management, Recycling and Conservation, Fourth Annual Report, University of Florida, Gainesville, 1978.
38. Boyt, F. L., S. E. Bayley, and J. Zoltek, Jr., Removal of Nutrients from Treated Municipal Wastewater by Wetland Vegetation. *J. Wat. Pollut. Con. Fed.* 49:789-99, 1977.
39. Odum, H. T., K. C. Ewel, J. W. Ordway, M. K. Johnston, and W. J. Mitsch, Cypress Wetlands for Water Management, Recycling and Conservation. Annual Report, University of Florida, Gainesville, 1974.
40. Kadlec, R. H., Pilot scale irrigation experiment - 1975; preliminary and partial results. In: The Effects of Sewage Effluent on Wetland Ecosystems, R. H. Kadlec, C. J. Richardson, and J. A. Kadlec, Editors. University of Michigan, Ann Arbor, 121, 1975.
41. Grant, R. R., Jr. and R. Patrick, Tinicum marsh as a water purifier. In: Two Studies of Tinicum Marsh. The Conservation Foundation, 1970.

42. Wellings, F. M., Interim progress report on effect of cypress domes, on viruses in effluent. In: Cypress Wetlands for Water Management, Recycling and Conservation, H. T. Odum, K. C. Ewel, J. W. Ordway, and M. K. Johnston, Editors. University of Florida, Gainesville, 802, 1975.

## ECOSYSTEM PROPERTIES RELEVANT TO ECOTOXICOLOGY

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A program for protecting ecosystems from the harmful impact of chemicals requires four kinds of investigation:

- (1) The search for general principles determining the relation between an ecosystem's structure and the impact on it of chemical input.
- (2) The description of particular ecosystems or classes of ecosystems to determine their structure.
- (3) The testing of individual chemicals for the ways in which they couple to the ecosystem and its components.
- (4) If the chemical is allowed into the environment, monitoring to verify predicted behavior or identify the unexpected.

The more completely we carry out (1) and (2), the better the prediction, the less stringent we have to be with (3), and the more efficiently we can carry out (4).

We would of course want to be able to extend the results of the study of a particular ecosystem to other similar ones. But, what do we mean by similar? It is not a matter of physical similarity, or having many species in common which justifies extrapolation. Therefore, we have to ask, what are the general properties of ecosystems which determine their qualitative behavior and which we should look at before predicting the impact? Or looked at the other way, what can make a system behave differently from predicted on the basis of extrapolating from another system?

The discussion below picks out 5 kinds of ecosystem properties which must be considered: random variation of parameters, arbitrary time dependent variation in the environment, the non-linear dynamics of the populations, network structure of the community, and natural selection within the component species.

### Connectivity

The outcome of a new input to an ecosystem depends not only on the component species but also on how they are linked together in networks.

Sometimes the graph of the ecosystem is sufficient to make qualitative predictions as to the direction of the effects.

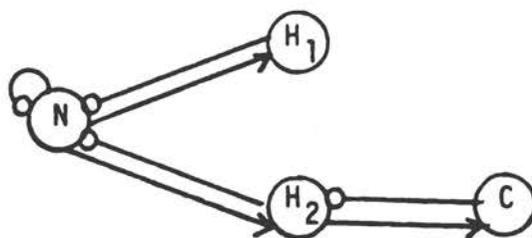
One such procedure makes use of signed graphs. It assumes that some parameters change slowly compared to the variables of interest, that these can track the parameter change. It asks, how will these variables change if they are tracking the parameters? It is necessary to specify the direction of effect of each variable on the others (that is, the sign of the first derivatives

$$\frac{\partial}{\partial x_j} \left( \frac{dx_i}{dt} \right)$$

This qualitative analysis is used in several ways:

(1) to establish general principles which suggest where to look for effects. For example, toxic impact concentrates upward in the trophic structure; or if the common resource of several species is reduced, the impact will be absorbed by the inedible specialized consumers. These generalizations are not universal but are a good guide as to where to begin to study.

(2) They provide a way of testing the proposed network models of an ecosystem. For instance, suppose that we recognize four components in a system: one nutrient resource, two consumers, and the predator of one of these. They are related by the graph



where  $\longrightarrow$  indicates a positive effect  
and  $\longrightarrow \circ$  a negative effect

But in addition there may be other interactions:  $H_1$  may inhibit  $H_2$  by some toxin,  $C$  may stimulate the growth of  $H_1$ , etc. If we do not know the structure of the network, we cannot predict impact. Therefore, we proceed by representing alternative models as shown below. For each one, we construct a table which indicates the direction of change of the variable listed above each column when the direct impact of a chemical enters the system as a positive input through the variable at the left of each row. (See Fig. 1)

This analysis shows that for many of the predictions the detailed structure doesn't matter. The predictions which coincide under different models are robust predictions. The places where the predictions of different models are disjunct are the places

to look to decide among them. The question marks indicate predictions which require measurement because different pathways have opposite effects.

(3) If we are not able to identify the source of an impact, we can still examine the correlations between variables. For example, in model 14, N and C respond in the same direction to impacts entering the system through  $H_1$  but in opposite directions to impacts entering through the other modes. Therefore, a positive correlation between N and C, and negative correlations between the variables and  $H_1$  and  $H_2$  identifies the source of the impact as  $H_1$ . In table 1, we show the pattern of correlations among variables for each model and for each input node. The 1's along the diagonal identify variables which change in either direction, and zero on the diagonal indicates variables which will not respond. The important point is that even though a prediction of +, 0, or - is a weak prediction, the joint confirmation of the correlation pattern can identify the source of the impact and sometimes can choose the best graph as well. Thus, an examination of the correlation pattern in an ecosystem is a strong test of its presumed structure, and therefore the basis for the prediction of impacts before they occur. It also allows us to answer how the system would respond to new variables entering the system, including the behavior of regulatory procedures.

These qualitative analyses may be sufficient to predict impacts. Or they may be used to decide which measurement to make in order to detect impacts.

### Random Processes

After we have made our best estimates of parameters in an ecosystem and derived our predictions from them, we have to recognize that our estimates may be in error and that the parameters vary over time. Therefore, the question arises, in what way might a system behave differently from expected if some of the constants turn out to be variables?

Variable parameters can be examined in two ways. First, they can be looked at as random variables and studied by the methods of stochastic equations. Usually, the randomness is treated as "white noise", with no correlation between the random variables at even arbitrarily close intervals. The results are expressed as probability distributions of population size, as probability of extinction, or as expected survival time to extinction. The second approach is to treat the parameter simply as some arbitrary time-dependent function.

The major results of the stochastic analysis are:

(1) Random variation due to fluctuations in parameters can cause the system to deviate from its expected behavior under constant conditions.

(2) Due to the multiplicative nature of population growth, the probability distribution of population size is often log-normal. That is, fluctuations may span several orders of magnitude, and the population may be far below its average value most of the time.

(3) Even when the expected behavior of a population would be to increase to some equilibrium level, random fluctuations can drive it to extinction.

(4) All of these behaviors increase with the variance of the parameter.

Therefore:

(a) A variable chemical input may increase the danger of extinction above what it would be with the same average value.

(b) A constant increased mortality may jeopardize a species which is fluctuating for other reasons. The "tolerable" mortality level is reduced by fluctuation.

(c) A chemical which increased the sensitivity of the organism to some variable factor in the environment increases the effective variance and therefore may threaten the species' persistence.

These are all conclusions derived from the dynamics of single species. They may be overridden in complex community interactions. At present, we do not have a satisfactory theory for random processes in ecosystems. Therefore, it is necessary to study such phenomena before we can predict what will happen.

### Nonlinear Responses

Until recently, ecological theory was dominated by models of linear interactions. If systems are near equilibrium (even if the equilibrium level itself is changing), this is not too serious a problem. But when systems are changing rapidly, new issues arise for which linear assumptions are inadequate.

The underlying processes linking variables in ecosystems are nonlinear in several ways. The response to increasing concentrations of a substance may level off as active sites saturate, or increase as detoxification mechanisms are saturable. A predator

may be satiated by increasing prey densities, or might be more effective in its predation when high densities of prey attract it. Mortality may decrease at high densities by saturating predators or increase by facilitating contagion.

Some of these processes, those most closely related to physiological kinetics, can be studied in the laboratory. But, those related to aggregative behavior, search efficiency, and dispersal of organisms are more difficult to reproduce in confined spaces and require more facilities or field study.

The nonlinear properties of interactions in the ecosystem are important both for the behavior of the system and for the research the system requires.

(1) If a response rate is a nonlinear function of some chemical's concentration or a population's density, testing must take place over a range of concentrations before predictions can be made.

(2) Most components and organisms have more than one possible fate. Whereas, in a linear system the partitioning is constant (a fixed proportion taking each pathway), in nonlinear systems, the relative importance of the pathways change with the concentration. A consumer with a high affinity and low saturation level may dominate the process at low concentrations and be completely overwhelmed at higher levels. While we can attempt to anticipate alternative pathways for laboratory experimentation, the possible occurrence of unexpected pathways which would alter the whole dynamics under some conditions urges the importance of experiments in more complex systems.

(3) The "constants" of the nonlinear kinetics (e.g., the affinity and asymptote of a Michaelis-Mention equation) may themselves depend on other components of the ecosystem.

(4) All habitats are heterogeneous. If a process responds linearly to concentrations and densities, its behavior over the whole system is its behavior at the average concentration. But, if the process is nonlinear, this is no longer true. A process which is an upwardly convex function of some component will be reduced by variable concentrations below what the average value of the concentration would suggest, while a concavely upward process is increased by variation. This means that in order to predict a response, we need to know how variable the concentrations or densities are in the habitat (which requires field study) as well as knowing the shape of the response function (which may be obtained in the laboratory in some cases).

(5) Even simple nonlinear systems are capable of showing many different kinds of behavior under constant conditions,

including alternative equilibria or sustained motion which may or may not be periodic. These non-equilibrium behaviors show regularities which require prolonged sequences of observations. Further, the systems may respond to toxic inputs differently at different points in their trajectories. This suggests that we determine when they will be most sensitive to damage, for instance by displacement from one mode of behavior to another.

It is possible to study the effects even of arbitrary, nonspecified variable parameters on ecosystems of given structure by the methods of time averaging. These techniques make use of the relation defining the time average or expected value of a function by

$$E_t(X) = \frac{1}{t} \int_0^t X(c) dc .$$

then 
$$E_t \left( \frac{dX}{dt} \right) = \frac{1}{t} (X(t) - X(0)) ,$$

and for bounded variables this goes to zero for long time intervals. For example, in the simple single-population model

$$\frac{dX}{dt} = rX(a(t) - X)$$

we can show that covariance  $(a, X) = \text{variance}(X)$  so that  $X$  is positively correlated with its food supply and varies less than  $a(t)$  does. Or if  $X$  feeds on species  $y$  at a rate that increases with  $y$ , say

$$\frac{dX}{dt} = X(y + y^2 - \theta)$$

for death rate  $\theta$ , we already know that

$$\bar{y} + \bar{y}^2 + \sigma_y^2 = \theta . \quad (\bar{y} \text{ is the average value of } y.)$$

therefore

$$\sigma_y^2 < \theta .$$

And if a change elsewhere in the system decreases the average  $y$  then the variance increases, making it more likely that  $y$  may become extinct. But if the relation were convex upward, say

$$\frac{dX}{dt} = X(y - y^2 - \theta) ,$$

the those factors which decrease  $\bar{y}$  also decrease  $\sigma_y^2$ . (This does not apply to changes in  $\theta$ ). Therefore, it is important to know if species respond in convex or concave ways to their prey or predators. Note here that regardless of the variability of environmental terms in the equations for other variables, the variance of  $y$  is controlled by  $\theta$ . It seems as if  $X$  will serve as a sink for variance in the system, absorbing increased variation.

It follows from this that before we can predict the consequences of a new input, we have to know the shape of the nonlinearities, which variables act as sinks for variance, which parameters control the variance.

### Natural Selection

Populations can adapt rapidly to changing conditions. The history of host changes in herbivorous insects, pesticide and antibiotic resistance, the overcoming of host resistance in fungi and insects and acclimation to new habitats suggest that such evolution may be very rapid. In the case of pesticides, 2-10 years is often sufficient for adaptation.

Adaptation by natural selection may be important in impact evaluation:

(1) Short term, pre-selection evaluation may overestimate the impact in the long run.

(2) Species are unequal in their capacity to respond. Those groups that can adapt quickly may come to dominate the biota. The difference between adapted and non-adapted species may override differential adaptation to other aspects of the environment and reduce diversity.

(3) The mode of adaptation may involve reduced growth rates, lower fecundity or stress tolerance, or altered food and habitat preferences which can change competitive relations.

The response to selection is proportional to the selection intensity and the additive genetic variance. Mortality tests in the lab could estimate genetic variance if the population sample is large enough, but effective selection pressure needs field study, and immigration from non-polluted habitats can swamp selection.

(4) Genetic changes (in the frequencies of biochemical markers) may be indicators of differential mortality or fecundity which might be difficult to evaluate otherwise. Similarly, observed response to selection can give a lower bound on selection intensity. And, if the age of the animal can be determined, the intensity of selection at each age class can be estimated. Such studies underestimate the mortality since we only pick up the differential rates, while population age structure overestimates toxicity since it registers all causes of death.

Proposal: For each habitat a set of species should be identified for which biochemical polymorphisms are known. Laboratory tests should search for differential mortality, and field study should monitor for genotype frequency change.

Table 1

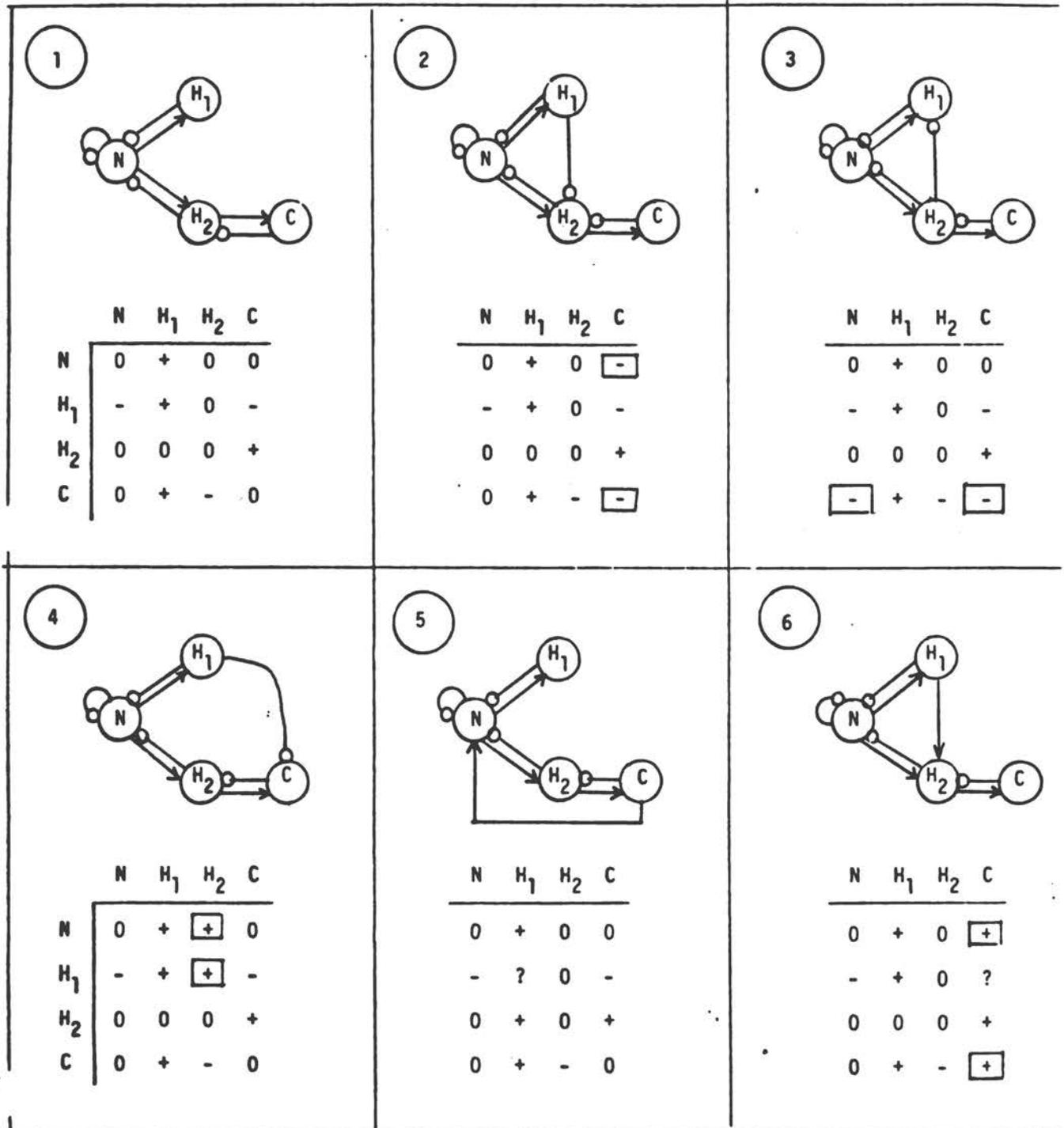
Model	<u>Input Correlations</u>																
	N				H <sub>1</sub>				H <sub>2</sub>				C				
	N	H <sub>1</sub>	H <sub>2</sub>	C	N	H <sub>1</sub>	H <sub>2</sub>	C	N	H <sub>1</sub>	H <sub>2</sub>	C	N	H <sub>1</sub>	H <sub>2</sub>	C	
1	N	0	0	0	0	1	-	0	+	0	0	0	0	0	0	0	0
	H <sub>1</sub>		1	0	0		1	0	-		0	0	0		1	-	0
	H <sub>2</sub>			0	0			0	0			0	0			1	0
	C				0				1				1				0
2	N	0	0	0	0	1	-	0	+	0	0	0	0	0	0	0	0
	H <sub>1</sub>		1	0	-		1	0	-		0	0	0		1	-	-
	H <sub>2</sub>			0	0			0	0			0	0			1	+
	C				1				1				1				1
3	N	0	0	0	0	1	-	0	+	0	0	0	0	1	-	+	+
	H <sub>1</sub>		1	0	0		1	0	-		0	0	0		1	-	-
	H <sub>2</sub>			0	0			0	0			0	0			1	+
	C				0				1				1				1
4	N	0	0	0	0	1	-	-	+	0	0	0	0	0	0	0	0
	H <sub>1</sub>		1	+	0		1	+	-		0	0	0		1	-	0
	H <sub>2</sub>			1	0			1	-			0	0			1	0
	C				0				1				1				0
5	N	0	0	0	0	1	-	0	+	0	0	0	0	0	0	0	0
	H <sub>1</sub>		1	0	0		1	0	-		1	0	+		1	-	0
	H <sub>2</sub>			0	0			0	0			0	0			1	0
	C				0				1				1				0
6	N	0	0	0	0	1	-	0	?	0	0	0	0	0	0	0	0
	H <sub>1</sub>		1	0	+		1	0	?		0	0	0		1	-	+
	H <sub>2</sub>			0	0			0	0			0	0			1	-
	C				1				1				1				1

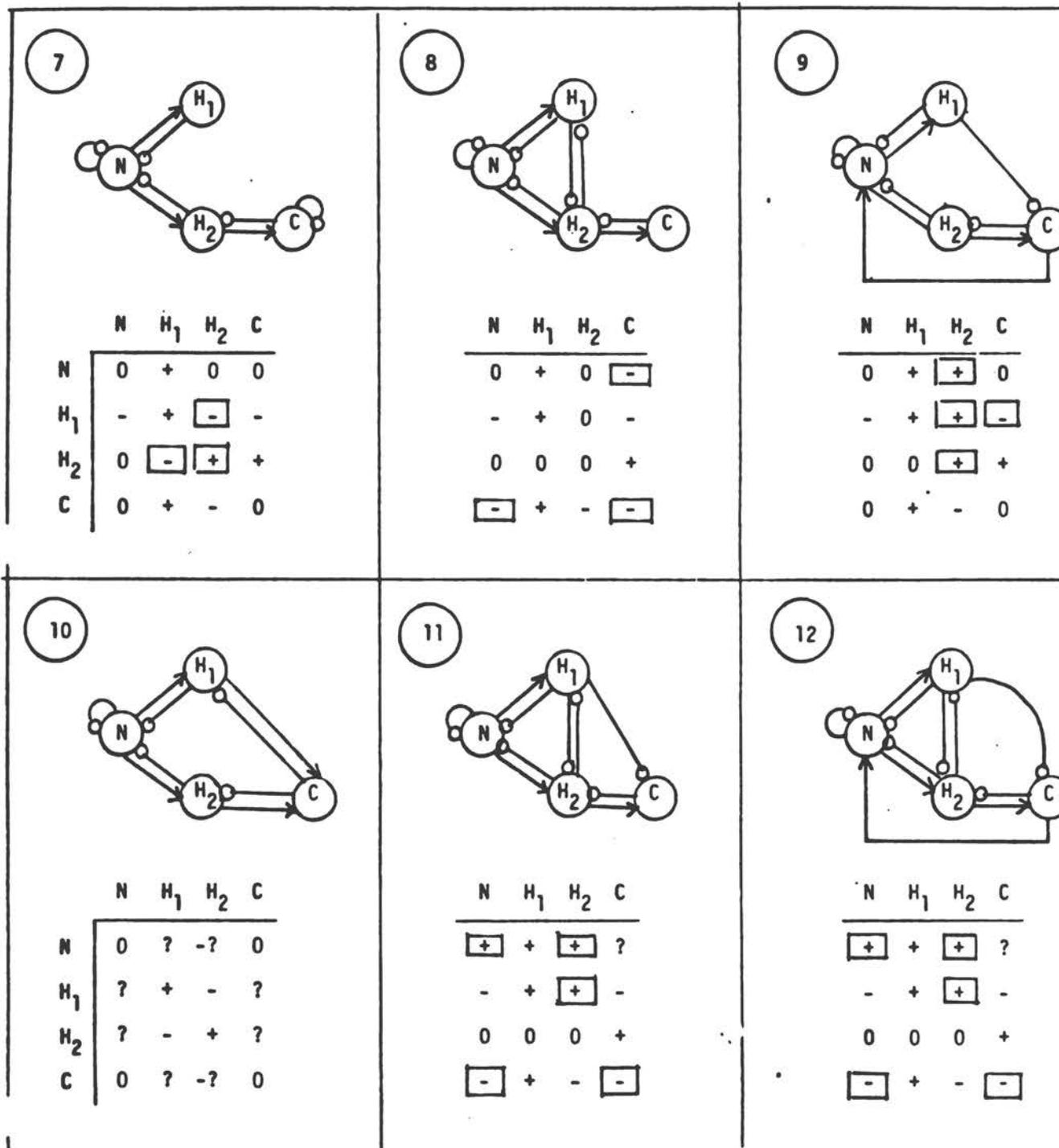
Model	N				H <sub>1</sub>				H <sub>2</sub>				C				
	N	H <sub>1</sub>	H <sub>2</sub>	C	N	H <sub>1</sub>	H <sub>2</sub>	C	N	H <sub>1</sub>	H <sub>2</sub>	C	N	H <sub>1</sub>	H <sub>2</sub>	C	
7	N	1	0	0	0	1	-	+	+	0	0	0	0	0	0	0	0
	H <sub>1</sub>		1	0	0		1	-	-		1	-	-		1	-	0
	H <sub>2</sub>			1	0			1	+			1	+			1	0
	C				0				1				1				0
8	N	0	0	0	0	1	-	0	+	0	0	0	0	1	-	+	+
	H <sub>1</sub>		1	0	-		1	0	-		0	0	0		1	-	-
	H <sub>2</sub>			0	0			0	0		0	0			1	+	
	C				1				1			1					1
9	N	0	0	0	0	1	-	-	+	0	0	0	0	0	0	0	0
	H <sub>1</sub>		1	+	0		1	+	-		0	0	0		1	-	0
	H <sub>2</sub>			1	0			1	-		1	+			1	0	
	C				0				1			1					0
10	N	0	0	0	0	1	a	-a	?	1	a	-a	?	0	0	0	0
	H <sub>1</sub>		1	-	0		1	-	?		1	-	b		1	-	0
	H <sub>2</sub>			1	0			1	?		1	-b			1	0	
	C				0				1			1					0
11,12	N	1	+	+	?	1	-	-	+	0	0	0	0	1	-	+	+
	H <sub>1</sub>		1	+	?		1	+	-		0	0	0		1	-	-
	H <sub>2</sub>			1	?			1	-		0	0			1	+	
	C				1				1			1					1
13	N	0	0	0	0	1	-	0	+	1	-	0	-	0	0	0	0
	H <sub>1</sub>		1	0	0		1	0	-		1	0	+		1	-	0
	H <sub>2</sub>			0	0			0	0		0	0			1	0	
	C				0				1			1					0

Model

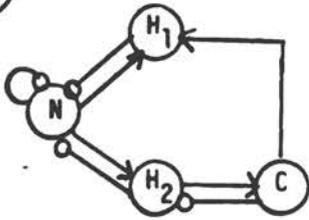
		N				H <sub>1</sub>				H <sub>2</sub>				C			
		N	H <sub>1</sub>	H <sub>2</sub>	C	N	H <sub>1</sub>	H <sub>2</sub>	C	N	H <sub>1</sub>	H <sub>2</sub>	C	N	H <sub>1</sub>	H <sub>2</sub>	C
14	N	1	+	+	-	1	-	-	+	1	-	-	-	1	+	-	-
	H <sub>1</sub>		1	+	-		1	+	-		1	+	+		1	-	-
	H <sub>2</sub>			1	-			1	-			1	+			1	+
	C				1				1				1				1
15	N	1	-	+	+	1	a	-a	-a	1	a	-a	-a	1	-	+	+
	H <sub>1</sub>		1	-	-		1	-	-		1	-	-		1	-	-
	H <sub>2</sub>			1	+			1	+			1	+			1	+
	C				1				1				1				1

Figure 1



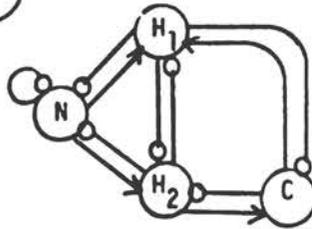


13



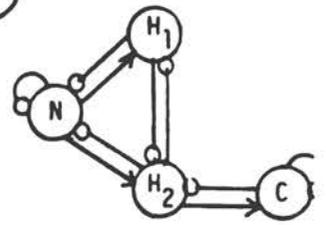
	N	H <sub>1</sub>	H <sub>2</sub>	C
N	0	+	0	0
H <sub>1</sub>	-	+	0	-
H <sub>2</sub>	-	+	0	+
C	0	+	-	0

14



	N	H <sub>1</sub>	H <sub>2</sub>	C
N	+	+	+	-
H <sub>1</sub>	-	+	+	-
H <sub>2</sub>	-	+	+	+
C	+	+	-	-

15



	N	H <sub>1</sub>	H <sub>2</sub>	C
N	-	+	-	-
H <sub>1</sub>	?	+	-	-
H <sub>2</sub>	?	-	+	+
C	-	+	-	-

# The Utility of Diatoms for Hazard Assessment of

## Chemicals in Ecosystems

Prepared for the National Research Council Commission on Natural Resources, Environmental Studies Board.

by

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Diatoms have been employed for assessment of environmental damage with much success over the past 30 to 40 years, Patrick, 1949, 1963, 1964. Most of this assessment has been accomplished by analyzing changes in diatom community structure. Changes in community structure, of course, merely represent a composite of changes of many species populations. It would seem then that by monitoring the populations of several critical species, one could assess potential environmental damage. This has been done with limited success in the diatoms. Kolkwitz and Marsson, 1908, were among the first to assign "Saprobien" tolerances to diatom species. These assignments were made from field observations rather than experimental manipulation and simply listed the tolerances of various diatom species to domestic sewage. Following this, several investigators have attempted to define the limits of tolerance of several diatom species to different potential perturbations. Weber and McFarland, 1972, reported the copper tolerance of several common lotic diatoms. Lanza and Cairns, 1972, investigated thermal stress on Navicula seminulum Hust. while Maloney and Palmer, 1956, studied the tolerance of several diatom species to six different chemical compounds. Most of the autecological data on diatom species, however, were generated from descriptive studies where investigators implied a species' tolerances based on distribution of that species. Most of this autecological information has been compiled for 300 common freshwater diatoms, Lowe, 1974. This

compilation considers species' responses to pH, nutrients, salt, organic pollutants (saprobien), current, habitat and temperature.

Although a great wealth of information has been generated concerning diatom requirements and tolerances, we do not know enough about any one species to use it alone in an autecological approach to hazard assessment. As new information is generated concerning diatom tolerances and assimilated into a compilation such as Lowe, 1974, diatom taxa have the potential of becoming excellent autecological indicators.

The state of the art today is the use of associations of species with "similar" ecological preferences, VanLandingham, 1974, Schoeman, 1976, 1979, Stoermer, 1978, Lange-Bertalot, 1978, 1979 and Descey, 1979. The changes in relative abundance among these associations indicate the general "health" of the stream. As our knowledge of diatom species ecology increases, we may be able to replace these indicator associations with indicator species but thus far we are left somewhere between a community and a species approach.

#### REFERENCES

- Descey, J. P. 1979. A new approach to water quality estimation using diatoms. Nova Hedwigia Beiheft 64: 305-323.
- Kolkwitz, R. and M. Marsson. 1908. "Ökologie der pflanzlichen Saprobien. Ber. Deut. Bot. Ges. 26: 505-519.
- Lange-Bertalot, V. H. 1978. Diatomeen-Differentialarten anstelle von Leitformen: ein geeineteres Kriterium der Gewässer belastung. Arch. Hydrobiol./Suppl. 51: 393-427.
- . 1979. Toleranzgrenzen und Populationsdynamik benthischer Diatomeen bei unterschiedlich starker Abwasserbelastung. Arch. Hydrobiol./Supple. 56: 184-219.
- Lanza, G. R. and J. Cairns. 1972. Physio-morphological effects of abrupt thermal stress on diatoms. Trans. Amer. Micros. Soc. 91(3): 276-298.

- Lowe, R. L. 1974. Environmental requirements and pollution tolerances of freshwater diatoms. EPA 670/4-74-005, 334 pp.
- Maloney, T. E. and C. M. Palmer. 1956. Toxicity of six chemical compounds to thirty cultures of algae. *Water and Sewage Works*. 509-513.
- Patrick, Ruth. 1949. A proposed biological measure of stream conditions based on a survey of the Conestoga basin, Lancaster Co., Pennsylvania. Proc. Acad. Nat. Sci. Phila. 101: 277-341.
- . 1963. The structure of diatom communities under varying ecological conditions. Ann. N.Y. Acad. Sci. 108(2): 359-365.
- . 1964. A discussion of natural and abnormal diatom communities. p. 185-204. In D. F. Jackson (ed.) *Algae and Man*. Plenum, NY.
- Schoeman, F. E. 1976. Diatom indicator groups in the assessment of water quality in the Jukskei-Crocodile River System (Transvaal, Republic of South Africa). J. Limnol. Soc. of South Africa, 2(1): 21-24.
- . 1979. Diatoms as indicator groups of water quality in the upper Hennops River (Transvaal, South Africa). J. Limnol. Soc. South Africa 5(2): 73-78.
- Stoermer, E. F. 1978. Phytoplankton assemblages as indicators of water quality in the Laurentian Great Lakes. Trans. Amer. Micros. Soc. 97(1): 2-16.
- VanLandingham, S. L. 1976. Comparative evaluation of water quality on the St. Joseph River (Michigan and Indiana, U.S.A.) by three methods of algal analysis. Hydrobiologia 48(2): 145-173.
- Weber, C. L. and B. H. McFarland. 1972. The effect of copper on the periphyton of a small calcareous stream. Unpublished manuscript presented at the 20th annual meeting of the Midwest Benthological Society, Ames, Iowa.



## ECOTOXICOLOGY AT THE WATERSHED LEVEL

by

Logan A. Norris<sup>1/</sup>

Assessments of environmental impacts from chemicals has traditionally been done at the single-organism level, usually using toxicology data from laboratory bioassay tests and estimates of exposure from laboratory or field studies of environmental chemistry. Relatively few tests have been done to assess the impacts of chemicals on single organisms in the field and few, if any, at the watershed (ecosystem) response level. There are two main reasons why there have been so few field tests: They are exceedingly difficult to do, and the current philosophies of hazard assessment have evolved from classical toxicology and the federal regulatory framework involving pharmaceuticals, food additives, pesticides, and the like. Techniques for evaluation of impacts of chemicals at the watershed level are needed because traditional methods of hazard assessment emphasize direct effects of chemicals on selected organisms. Indirect effects may be of equal or greater importance in maintaining ecosystem structure and function.

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## DIRECT AND INDIRECT EFFECTS

Direct effects of chemicals on organisms are those that result from the direct contact of a specific organism with a chemical agent. These effects require that a sequence of events occur: (1) The chemical and the organism must come in contact, (2) the chemical must be taken up by the organism, (3) the chemical must move from the point of uptake to the point of biochemical action, and (4) enough chemical must be at the site of biochemical action in an active form for a sufficient period of time that a biochemical effect can take place. Toxicity results from the direct interaction between an organism and a chemical. The nature of the toxic response depends on the basic properties of the chemical and organism and the kind of exposure the organism receives (i.e., the magnitude, frequency, duration, and route of exposure). Direct chemical effects can be studied and evaluated in terms of currently accepted dose-response theory.

An ecosystem implies a collection of different kinds of organisms among various kinds of nonliving components. The theories pertinent to direct chemical effects on one organism are perfectly applicable to all organisms in an ecosystem, because the direct interaction between the organism and the chemical is a one-on-one process. From the standpoint of dose-response relationships, it is immaterial whether the 50 mg/kg at the site of biochemical action came from inhalation, ingestion of contaminated water, or consumption of other organisms. The difficulty is that an ecosystem may have an extremely large number of different

kinds of organisms, most of which are not likely to be involved in traditional programs of toxicological testing. Representative species of major groups of organisms may be selected to predict direct chemical effects, but they may not in fact reflect the reaction of some particularly important organism in the ecosystem. For instance, the species selected for study could be much more or much less sensitive than most of the species of the class of organisms that the test species is meant to represent. In this case, the direct effects of the chemical will be over- or underestimated. Through study and careful selection of test species, however, much can be estimated about direct toxic effects through classical dose-response experimentation. It is necessary, however, to assess the consequences to the ecosystem which accrue from changes in the numbers or activities of all the directly affected organisms.

Indirect effects are those which do not require a direct interaction between the chemical and an organism. As an example, a chemical which reduces the primary productivity of an ecosystem may not have a direct toxic effect on organisms at higher trophic levels; but the reduction in primary production may have an overwhelming effect on these organisms. The actual event which caused the reduction in primary production is less important than the occurrence of the reduction. In this example, the effect on higher trophic levels results from the reduction in primary production—not interaction with the chemical.

In studying events of this kind, then, it may be possible to examine the processes involved and to manipulate them by a variety of techniques,

some of which may not be related at all to the chemical in question. If the techniques produce the same effects on primary productivity as does the chemical, they are useful for studying the effects of reduced primary production.

In many cases, the indirect effects may be substantially more far-reaching (in an ecosystem perspective) than direct chemical effects. Most chemicals are not likely to directly affect all organisms in any ecosystem simply because few compounds are so inherently toxic or so widely distributed that the avoidance or detoxification mechanisms of all organisms would be overwhelmed. On the other hand, severe deleterious effects on only a few key organisms can have far-reaching indirect effects for all other components of the ecosystem.

#### PERSPECTIVES ON PERTURBATIONS

Chemicals are only one of a wide variety of agents or activities that can and do alter the structure and function of ecosystems. If chemicals are unique, it is because their direct effects may be less likely to affect the wide range of organisms that other perturbations do. For instance, logging and burning slash in forested watersheds, tilling in agricultural watersheds, and impoundments in aquatic systems are all perturbations that have direct effects (as well as many indirect effects) on a wide range of organisms. The methods which have been used in studying the effects which result from these events are the same as those that can be used to study any perturbation, whether it is initiated

by a chemical or some other event. The key is to recognize that when chemicals are involved, the direct effects can be studied from a dose-response perspective including, but not limited to, single species studies.

#### INFORMATION NEEDED TO ASSESS EFFECTS OF CHEMICALS AT THE WATERSHED LEVEL OF RESOLUTION

Watersheds represent an amalgamation of an extremely diverse and complex series of subsets of systems which interact among themselves. There are some major processes, however, which involve or affect all or most of the subsets and which can be studied or measured as indicators of change in watershed-level ecosystems. These processes include carbon fixation by primary producers, transfers of energy, nutrient cycling, and the decomposition of all kinds of organic substrates.

The processes outlined above are often measured in watershed-level, ecosystem studies. Of course, the methods of study require sampling as a basis for expanding point measurements to the watershed level. Watersheds are physically defined by their hydrologic boundaries. In this sense, it is the hydrologic-related or mediated events or processes that are measured directly at the watershed level. The quantitative and qualitative aspects of water, nutrient, and sediment discharge are the direct measures of "watershed-level" activities.

All the processes included in this section reflect the activities of a wide range of organisms. Therefore, the values measured represent

some integration of the individual contributions of each participant. It is entirely possible that changes in the level of activity in one of these processes by any one organism could be offset by the action of another organism such that no net effect would be observed at the watershed level of resolution. If this type of resiliency exists (either from compensation or recovery), then it may be appropriate to conclude that significant ecotoxic effects were not apparent at the watershed level of resolution even though they may have been occurring at the species level.

To determine if changes are occurring in the processes or responses outlined above, it is necessary to have baseline values, including a measure of the variation of the system as a function of time. The use of paired watersheds or areas (control and treated) will be necessary. Unfortunately there are few watersheds that can be used in assessments of this kind. The number of such watersheds is vanishingly small compared to the number of chemicals (even groups of chemicals) covered by the Toxic Substances Control Act (TSCA). Watershed-level studies will most likely have to be part of a research rather than a regulatory program. This research, however, needs to be done to establish the linkages or relationships between small-scale, process-level studies (like carbon and nitrogen fixation, etc.) and the watershed-level response studies of water, nutrient, and sediment yield.

## FACTORS AFFECTING THE BEHAVIOR OF CHEMICALS IN WATERSHEDS

The behavior of a chemical includes its movement, persistence, and ultimate fate in the system. Behavior of a chemical determines organism exposure, because behavior determines how much chemical is where, for what period of time, and in what form. Behavior is, therefore, of equal importance with toxicity in determining the likelihood that direct chemical effects will occur in organisms. Behavior, that is, exposure, may be viewed as the "dose" just as toxicity characteristics may be viewed as the "response" in dose-response relationships.

A basic tenet of environmental chemistry (or chemodynamics) is that the properties of a chemical in the environment interact with the properties of the environment to produce the particular movement, persistence, and fate of the chemical. The properties of the chemical which are most important in determining behavior are: (1) Water solubility, (2) equilibrium vapor pressure, (3) partition coefficient, and (4) pK. The properties of the environment, which are important in determining chemical behavior, are numerous and subject to substantial variability across the range of conditions found in any one watershed. The most important properties are (A) climatic (radiation, temperature, precipitation, and wind), (B) edaphic (the physical and chemical properties and depth of the soil, the characteristics of the litter layer and soil organic matter, physical-chemical properties of the aquatic zone), (C) topographic (slope, aspect, and elevation), and (D) biotic (species composition and density of the vegetative and the soil microbiological communities).

These are the factors which will influence the behavior of chemicals in the environment. Knowledge of the important characteristics of both the chemical and the environment will provide a reasonable basis for making first predictions of chemical behavior in a particular environment. It is important to recognize, however, that our ability to predict chemical behavior is quite rudimentary (although a number of models of chemical behavior have been developed).

#### CHEMICALS OF CONCERN

A huge number of chemicals are covered by the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) and TSCA, and their properties are as diverse as the properties of ecosystems. A few generalizations and perspectives, however, may be helpful. A list of characteristics of chemicals which are important will be more helpful than a listing of chemical groups because of the diversity of properties within groups of chemicals. The following is a description of chemical-physical properties or characteristics of environmental behavior that will help guide attention to specific chemicals which are more likely to cause changes in ecosystems at the watershed level of resolution.

Heavy metals or those compounds that contain heavy metals.—These materials are characteristically persistent, chronically toxic, and accumulate in both the physical and biological portions of the environment.

Low water and high fat solubility.--These properties favor bioaccumulation and may favor the extensive adsorption of chemicals by sediment in the aquatic zone. These are both characteristics which may result in substantial and continuing exposure of organisms and a potential for increased exposure at higher trophic levels.

High equilibrium vapor pressure.--This characteristic favors vaporization and therefore rapid distribution of compounds across the ecosystem. On the other hand, it may also result in the rapid removal of the chemical even though this may only represent transport of the chemical from one location to another.

High degree of stability.--Some types of chemical structures (like those in PCB, DDT, and TCDD) are highly resistant to physical, chemical, or biological degradation. This may result in long persistence in the environment and a greater probability of redistribution and organism exposure. Compounds with this property and which are chronically toxic need special attention.

High degree of mobility in soil.--This characteristic may favor groundwater pollution, particularly if the compound is highly mobile and also highly persistent.

## FEATURES OF WATERSHEDS WHICH INFLUENCE TOXICITY

There are numerous features of watersheds that can influence both direct and indirect effects of chemical insult. These features were discussed in sections above. In general, the direct effects from a given chemical will be influenced by any factor which influences organism exposure. This means those factors which influence the movement, persistence, availability, or fate of the chemical in the environment will influence the degree to which direct chemical effects are likely to occur. Thus, in a cold, dry, nutrient-poor environment which reduces the rate of chemical or biological degradation, organisms may receive substantially greater exposure. The probability of groundwater pollution from chemicals which are both persistent and mobile is greater in environments with high precipitation and coarse-textured soils with low organic matter. On the other hand, organism exposure (and therefore the likelihood of toxic effects occurring) is substantially reduced in watersheds which are warm, moist, nutrient-rich, and highly active biologically and have abundant organic matter, gentle slopes, and deep soils.

Indirect effects are mostly influenced by the resiliency of the system, both with respect to a systems ability to compensate and/or recover from the alteration of a critical process. Ecosystems which are of limited diversity and only marginally stable (perhaps because of previous perturbations or harsh environments) are much more susceptible to a wide range of indirect effects than systems which have greater

diversity and stability and thus a greater ability to compensate and/or recover from a disturbance.

## REFERENCES

Bormann, F. H., and G. E. Likens.

1979. Pattern and process in a forested ecosystem. 253 p. Springer-Verlag, New York.

Franklin, Jerry F., L. J. Dempster, and Richard J. Waring, eds.

1972. Proceedings - research on coniferous forest ecosystems - a symposium. [Bellingham, Wash., March 1972] 323 p. Pac. Northwest For. and Range Exp. Stn., Portland, Oreg.

Hall, Charles A. S., and John W. Day, Jr., eds.

1977. Ecosystem modeling in theory and practice - An introduction with case histories. 684 p. John Wiley & Sons, Inc., New York.

Haque, Rizwanul, and V. H. Freed, eds.

1975. Environmental dynamics of pesticides. 387 p. Plenum Press, New York.

Innis, George S., ed.

1978. Grassland Simulation Model. 298 p. Springer-Verlag, New York.

Knisel, Walter G., Jr.

1978. A system of models for evaluating non-point-source pollution-- an overview. CP-78-11. International Institute for Applied Systems Analysis, A-2361 Laxenburg, Austria.

Leonard, R. A., G. W. Langdale, and W. G. Fleming.

1979. Herbicide runoff from upland piedmont watersheds--data and implications for modeling pesticide transport. *J. Environ. Qual.* 8(2):223.

Tinsley, Ian J.

1979. *Chemical concepts in pollutant behavior.* 265 p. John Wiley & Sons, New York.

Webb, Warren L., Henry J. Schroeder, Jr., and Logan A. Norris.

1975. Pesticide residue dynamics in a forest ecosystem: a compartment model. *Simulation* 24(6):161.

THE UTILITY OF SINGLE SPECIES  
AND ECOSYSTEM TESTS IN ASSESSING  
THE ENVIRONMENTAL IMPACT OF RADIONUCLIDE ECOTOXICANTS

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## INTRODUCTION

Radionuclides as a class of ecotoxicants differ in many ways from other ecotoxicants and possess a large variety of physical and chemical properties. Radionuclides have a variety of finite lifetimes such that they essentially disappear over time. Some fission product radionuclides including iodine-131, strontium-89, and ruthenium-103, have half-lives measured in days. Others, such as tritium (hydrogen-3), strontium-90, cesium-134, cesium-137 and isotopes of plutonium, americium, and curium have half-lives measured in years to centuries. Radionuclear ecotoxicants, such as krypton-85, may be biologically inert, or they may be biologically active as is the case for C-14 or tritium. Radionuclides such as cesium-137 and cobalt-60, enter the sedimentary chemical cycle whereas others, such as C-14 and tritium, may cycle between gaseous and other states of terrestrial or aquatic ecosystems. A further difficulty in categorizing the radionuclear ecotoxicants is the nature of their emissions - alpha, beta, or gamma-ray - which differ significantly both with respect to their potential as a hazard to life and the radiation fields they create. Radionuclear materials thus represent a diverse array of ecotoxicants not easily categorized.

The study of radionuclear exotoxicants spans a period of over 80 years and significant environmental research began over 30 years ago. In the course of this research, both laboratory and field experimentation have been conducted to determine the biological effects of ionizing radiation exposures. Similarly the dynamics of radionuclear materials in biogeochemical systems have been evaluated in microcosms under laboratory conditions and in ecosystems under naturally-occurring field conditions. Laboratory experimentation has been characterized by single species testing to determine the effects of a variety of ionizing radiations. The use of

both single species and ecosystem testing throughout the comparatively long history of radionuclear ecotoxicant research allows for comparison of these two methods of assessing the environmental impacts of ecotoxicants.

This paper addresses the utility of single-species tests for assaying impacts of radionuclear ecotoxicants by examining the sufficiency of the single species method. For purposes of this paper, single species tests will refer to impact assessments using either single species or synthetic laboratory microcosms which are mixtures of limited physical and biological components of an ecosystem. Among the advantages of such single species assessments are 1) a well-defined system, 2) measurable and repeatable parameters, 3) relatively rapid response, and 4) evaluation of a direct effect. Some disadvantages of using the single species approach are 1) single species responses are buffered by ecosystem phenomena, 2) species interaction effects are excluded, 3) lack of assessment of emergent system properties and 4) failure to account for ecosystem compensation of damage to a single species. Ideally, evaluation of ecotoxicant impact by single species analysis should lead to the same conclusions derived from ecosystem analysis. Since the ecosystem approach is the meterstick for comparison, the sufficiency of the single species method can be determined by the extent of agreement between conclusions derived from single-species and ecosystem approaches.

#### MODES OF ENTRY

Radionuclides in a gaseous, aerosol, particulate or ionic state enter specific ecological systems through atmospheric, aquatic, or terrestrial pathways from a diversity of radionuclear sources. Atmospheric testing of nuclear weapons during the decade of the 50's initiated worldwide distribution of radionuclides which subsequently were deposited on ecosystems

throughout the world. Commercial nuclear fission reactors and associated nuclear fuel cycle activities are continuing sources of radionuclide release to the atmosphere along with occasional atmospheric detonations from nuclear weapons testing by nations not participating in the nuclear test ban treaty. The atmospheric release of radionuclides are considered global level environmental events since the radionuclear materials injected into the atmosphere may be transported through the global atmosphere subject to removal by rainfall, snowfall, and direct impaction on surfaces (Klement 1965).

In contrast to the global nature of atmospherically-released radionuclides, releases of radioactivity to freshwater aquatic ecosystems are of local impact since the radionuclear materials are contained in a restricted medium. Aquatic releases of radioactivity occur from a variety of military, governmental, industrial, medical, and educational sources. Non-routine fresh water releases of radionuclides by nuclear facilities may occur as a result of emergency needs for large quantities of water. Radionuclear releases to marine ecosystems may be considered both local and global in nature. The development of nuclear, ocean-going vessels has provided a direct source of radionuclide release both at harbors and over the oceanic routes followed by these vessels. Also, radionuclear materials from fission reactors and reprocessing facilities are released to oceans by some nations. (Radioactive effluents from nuclear fuel reprocessing plants 1978).

The third major mode of entry of radionuclides into ecosystems is through burial and storage of low level and high level radionuclear wastes. Low level wastes are routinely buried in shallow pits by facilities handling radionuclear materials or by a commercial low level

burial facility. Currently, high level wastes are temporarily stored awaiting development of a National Repository for long-term storage.

Long-term storage of high-level radionuclear wastes will probably be deep burial in isolated geological formations. The potential entry to ecosystems of radionuclides buried deep in the earth differs significantly from other modes of entry in a number of important respects. The time considerations for radionuclear decay and possible transport are on the order of hundreds to thousands of years. Although a deep burial site has not been selected, major problems associated with radionuclear containment are primarily those of potential long term release to groundwater systems and subsequent emergence of radioactivity in rivers or through pumping of groundwater (Waste Isolation Safety Assessment Program Technical Progress Report for FY-1978,1979). Another major source of repository release could be by accidental or purposeful human intrusion of the repository through mining or drilling for non-nuclear materials.

#### POINTS OF IMPACT

The modes of entry of radionuclides into terrestrial, fresh water, and marine ecosystems can be categorized as release from a concentrated source and subsequent dilution within a larger environment. Two central considerations emerge when radioactive materials have entered a specific ecosystem. First, what is the immediate or short-term impact of the radioactivity as it reaches the biota of the ecosystem? Second, what is the longer-term or chronic impact and fate of the radioactivity? The second consideration introduces considerable complexity into radionuclear impact analysis since radionuclides may be reconcentrated by biota or in physical components due to the dynamic nature of physical, chemical and biological processes of an ecosystem. The utility of single-species and ecosystem

approaches to assessment of immediate radiation impacts is discussed under the section: ionizing radiation fields. A similar comparison for the long-term fate of radioactivity is considered under the topic: radionuclide cycling and transport.

Ionizing Radiation Fields - An enormous number of experiments have been conducted to assay the sensitivity of plant and animal species to ionizing radiation. The majority of these studies were conducted using X-ray and gamma-ray radiation sources to which the organisms were either chronically or acutely exposed. Such studies have documented great differences in both animal and plant species sensitivities to ionizing radiation (Bond 1969, Wegender 1966, Sparrow 1966, Sparrow et al. 1968, Gunkel and Sparrow 1961). Research with plants (Sparrow 1963, Sparrow et al. 1965, Witherspoon 1967) led to predicative relationships between nuclear volume-chromosome number and radiosensitivity for ionizing radiation effects on species not previously tested and provided greater understanding of the mechanisms of damage.

Parallel measurements of ionizing radiation effects on plants in ecosystems have shown that a predicted lethal dose does not always produce plant mortality. For example, Chappell (1963) found that Smilax spp. (greenbriar) was afforded special protection from lethal radiation doses due to its underground vegetative structure which allowed emergence of new above-ground shoots. Ragsdale and Rhoads (1974) reported a similar result for Larrea divaricata shrubs following lethal doses to above-ground stems. Larrea recovered from the lethal exposure due to a capacity to generate new above-ground shoots from basal or subterranean tissue. Other shrubs associated with Larrea and receiving lethal radiation doses did not recover. Ecosystem tests of plant radiosensitivity showed that the

capacity for basal or subterranean sprouting is a significant factor in evaluating species radiosensitivity in an ecological system.

The sensitivity of an organism to ionizing radiation may vary with the life-stage of the organism. The seed of deciduous trees are much more resistant to ionizing radiation during dormancy than just after seed dormancy is broken (Heaslip 1959). Variable life-stage radiosensitivity and the effects of irradiation methods have been shown in studies with tree seed production (Witherspoon 1968). The internal dose from cesium-137 within the plant tissues decreased seed production 10 times below that reported for externally irradiated plant reproductive tissue. Differential life-stage radiosensitivity was shown for both irradiation methods since radiation doses causing seed production damage were much lower than those required to cause visible damage or mortality to growing plants. The eggs of the fish, Cyprinus carpio (carp), display variable radiation sensitivity during the period from 30 minutes following fertilization to 24 hours (Frank and Blaylock 1971). Based on carp egg hatchability, the most radiosensitive stage is at 30 minutes following fertilization, the next most sensitive stage is at 3 hours post-fertilization and radioresistance increases as egg development continues. Differences in life-stage radiation sensitivity are known for a variety of aquatic organisms for which the most general observation is that earlier life-stages are more sensitive than later life-stages (Polikarpov 1966). Insects exhibit differential life-stage radiosensitivity with greater sensitivity usually associated with younger life-stages (Menhenick and Crossley 1968, 1969). Further, radiosensitivity of a given insect life-stage apparently varies with overall culture conditions. For example, irradiated crickets of the genus Acheta were found to suffer a greater mortality under field conditions than in the laboratory as a result of greater predator and pathogen pressure

under field conditions (Auerbach, Crossley and Shinn 1968).

The effects of gamma and neutron radiation on whole intact ecosystems has been studied by a number of researchers (Woodwell 1962; Platt 1963, 1965; McCormick 1963, 1967; McCormick and McJunken 1965; Witherspoon 1965; Cotter and McGinnis 1965; Woodwell and Rebeck 1967; Woodwell and Holt 1971; Zavitkovski and Rudolph 1971; Fraley and Wicker 1973). These researchers and others documented unique plant effects including greater sensitivity of terminal buds, delayed plant dormancy, direct regrowth of trees by basal sprouting, delay of radiation damage from one growing season to the next, and the significance of environmental conditions in altering plant response to a given radiation dose.

The most general result from the forest ecosystem studies was that radiation affected the structure of the forest community by removing the canopy dominants and re-initiating succession at the herbaceous level. This produced a number of ecological impacts including lowered productivity, reduced species diversity, loss of nutrient elements as a result of lowered biotic retention capacity, and reduction of food web complexity. As a result of differential sensitivity of trees, radiation acted as a selective removal agent capable of altering species diversity and forest composition over long time periods. In some cases, differential radiosensitivity of species resulted in competitive interaction which further changed species distributions beyond that caused directly by radiation.

Following almost a decade-long period of research with gamma or neutron radiation in single species and ecosystem assessment, ecosystem studies at the Nuclear Test Site in Nevada provided new results. In the analysis of desert shrub damage from natural fallout fields around the Plowshare underground nuclear tests (Rhoads et al. 1971), it was shown that the beta component of the radiation dose was far more prevalent than

the gamma component. The beta to gamma-ray dose ratios were on the order of 5 to 14 and most of the radiation dose in areas where vegetation was damaged was attributable to beta radiation. The difference between beta and gamma fields is significant since, as a result of gamma-ray penetration, gamma-ray doses to vegetation accrue from relatively large fallout areas; whereas beta doses to vegetation are a function of particle proximity to the tissue and local beta radiation fields. The beta radiation resulted in differential damage or death to the shrubs which served as fallout particle filters. Through time the beta radiation field over the landscape became non-uniform and higher beta doses were associated with vegetation where radioactive particles accumulated. Murphy and McCormick (1971) found that fallout beta radiation doses producing 18 to 50% bud mortality had little overall effect on subsequent plant height, leaf length-width ratio and overall biomass.

Comparison of the results of species and ecosystem tests of the radiosensitivity of organisms shows that each has an appropriate use. At the single species level useful predictive relationships between radiation dose and biotic damage were formulated on fundamental measurements of nuclear volume and chromosome number. The single species approach provided a baseline of potential damage to life and also contributed to understanding of damage mechanisms. However, as clearly shown by ecosystem studies, predictions based on single species tests were subject to error. Under ecosystem testing, some organisms died when they received a lethal dose and other organisms survived simply because of their asexual reproductive capacities. Hence, it was not possible to predict which organisms would be most severely impacted based on single-species tests under laboratory conditions. Ecosystem testing revealed a number of plant responses which were not, or could not have been, elicited from single species tests.

Further, the ecosystem tests revealed the nature and extent of radiation damage as it impacted a functioning ecosystem. Neither the direct radiation effects nor the subsequent effects related to species interactions could have been predicted from single species testing.

A potentially more serious error arising from the single species approach was a general failure to recognize the enormous importance of the beta component of ionizing radiation in evaluating radiation impacts. Although beta radiation has now been studied at the single species and ecosystem levels, the knowledge of the significance of beta radiation arose from ecosystem testing and analysis of radiation effects. Miller (1971) in addressing the issue stated that the nature of the hazard of gamma-rays and the response of biological species to it are better known than for the other radiation hazards.

Radionuclide Cycling and Transport - Species-level laboratory testing of radionuclide cycling has been performed for both aquatic and terrestrial systems through the use of microcosms. Microcosms simulate larger natural systems and allow investigator manipulation of the physical and biological components of the system. Witkamp and Frank (1967) followed cesium-137 flux in terrestrial microcosms. Patten and Witkamp (1967) simulated the flux of radiocesium in terrestrial microcosms of varying levels of biotic and abiotic complexity. Ragsdale, et al. (1968) analyzed the effects of radiation on cobalt-60 and cesium-137 dynamics of aquatic microcosms of varying complexity. Witkamp and Merchant (1971) analyzed the effects of physical and nutrient variables on radionuclide distribution in producer-consumer microcosms. Microcosm studies such as these illustrate that the radionuclear concentrations of organisms are extremely sensitive to the nature of the microcosm. The complexity of the microcosm affects the flow-through or turnover of the radionuclide. Further, where

soil is important as a sink or regulator, the choice of soil substrate can have a significant effect on interpretation of the impact of specific radionuclides (Tamura and Jacobs 1961). Microcosms have been exceedingly useful in illustrating that the flux pathways are highly dependent on the nature of coupling of species in trophic relations and in illustrating effects of the biotic component on the physical substrate with respect to cycling and accumulation.

Ecosystem studies of radionuclide cycling in the field have provided the bulk of our knowledge of the flux of radionuclides and their fate in the environment. Early in the history of fallout studies, the need for information from the natural landscape was recognized. The first experimental study of radionuclear material cycling began following Project Trinity, a nuclear detonation at the Nevada Test Site in 1947 (Larson 1963). Subsequently, field ecological studies were initiated in a number of biomes (Larson 1971). Early reports of bioaccumulation of a fission nuclide resulted from studies in the Arctic region (Hanson and Palmer 1965, Hanson 1967) in which cesium-137 bioaccumulation through the lichen, caribou and Eskimo food chain were documented. Abrupt seasonal increases of cesium-137 in caribou were found to be related to seasonal radiocesium cycling in lichens (Hanson and Eberhardt 1971). Other researchers (Holleman and Luick 1975) working with body burden peaks of radiocesium in reindeer demonstrated that increases in the potassium concentration of the food of reindeer produced a 2-fold decline in cesium-137 concentration. A seasonal shift in reindeer diet from low potassium-high cesium-137 lichens to high potassium leafy green vegetation was postulated as the mechanism responsible for annual summer declines in reindeer whole body burdens in cesium-137.

Radiocesium bioaccumulation was found for deer in the United States Coastal Plain (Jenkins and Fendley 1971). Contrary to the then existing concerns over radiostrontium biomagnification, it was found that fallout cesium-137 was biomagnified by a factor of 3 in white tailed deer of the Coastal Plain physiographic province. Biomagnification of cesium-137 was also found for other game animals of southeastern Coastal Plain (Jenkins, Monroe and Golley 1967 and Jenkins and Fendley 1968). The explanation for this biomagnification is complex involving nutrient conditions, soil-fixing capacity, water table heights, and dietary items. Gamble (1971) in explaining why cesium-137 found in Florida milk was 6 times higher than the national average proposed a cesium-137 recycling mechanism involving direct return of radiocesium from litter to root by mycorrhizae. Dahlman, Francis and Tamura (1975) in reviewing cesium-137 cycling in terrestrial habitats concluded that atmospheric deposition and increased plant uptake could explain cesium-137 concentration in Coastal Plain plants.

Radiouclear ecotoxicants with long half-lives not only recycle within the ecosystem in which they are deposited, they also undergo long-term flow between adjacent landscape ecosystems. Ragsdale and Shure (1973) working in a cesium-137 effluent stream documented the significance of lateral redistribution processes across swampy flood plains and the long term consequences of the process for radiocesium cycling. Others working in the same and similarly-contaminated streams (Shure and Gottschalk 1975, 1978; Hay and Ragsdale 1978; Sharitz, et al. 1975; Garten, et al. 1975; Gladden 1979) have documented cesium-137 uptake, recycling, and bioaccumulation. Gladden (1979) summarized the myriad processes and interactions relating to radiocesium transfers among stream-bank-flood plain zones and along the downstream gradient.

Similar landscape scale movement of radionuclear ecotoxicants have been documented for terrestrial ecosystems. Ritchie, et al. (1980) found cesium-137 accumulating in a marsh receiving erosional runoff from agricultural fields within the watershed. The marsh served as a particulate filter accumulating cesium-137 from the runoff.

The comparison of single-species or microcosm assessment to ecosystem assessments clearly demonstrates the insufficiency of the single species microcosm approach as a simulator of ecosystems. For example, the biomagnification of cesium-137 in the Arctic food chain and in the Coastal Plain deer and higher trophic level vertebrates might not have been predicted since it is unlikely, due to the size of the organisms and the subsequent cost of the research, that these organisms would have been used in single species tests. It is equally unlikely that the exact chemical element environment of caribou, deer or other vertebrates could have been duplicated even if these organisms were the subject of single species tests. We have known since the early 60's that sandy soils had much lower retentive capacity for radionuclides than the clay soils. However, that single species level knowledge was not sufficient to predict the subsequent bioamplification of cesium-137 in organisms of the Coastal Plain. The possible mychorrhizal role in direct cesium-137 transfer to roots and the effect of naturally-changing nutrient status on cesium-137 flux could not have been predicted from microcosm studies since at that time the mychorrhizal relationship was not well enough known to be included in such tests. Even today the inclusion of mychorrhizae in microcosm tests may require more time and greater cost than can be allocated for it. It is virtually self-evident, but none the less true, that accumulation and reconcentration of radionuclear materials through major landscape flows

between ecosystems could not be predicted from single species microcosm testing.

While the comparison of microcosms and landscape studies illustrates in a vivid way the insufficiency of microcosm testing, the comparison also reveals limited uses for the microcosm approach. Single species tests are useful in preliminary trials prior to large scale ecological studies, they are useful for study of specific mechanisms identified from landscape studies, and their utility in theoretical research is well established. Species level research which includes single species testing, microcosms, and limited field experiments can explain phenomena observed at the ecosystem level. However, species level research cannot be used to project ecosystem phenomena.

#### TEMPORALITY

Correlation of change in radionuclear concentration with season has been documented in a variety of ways. Fallout arrival from the atmosphere to the earth's surface is a function of spring injection of fallout and subsequent precipitation. Hence, in areas with seasonal rain or snowfall, the influx of radioactive fallout will be seasonal. In geographical areas with annual leaf fall, a relatively high influx of radionuclear materials to the forest floor will occur during the fall months. Concentrations of radiocesium in woody plants are normally highest for reproductive tissues and young leaves during the early spring in the Coastal Plain. The seasonality of peak whole body cesium-137 burdens of caribou, reindeer and Coastal Plain deer was described above. Seasonal variation in radiocesium content was reported by Garten, et al. (1975) with changes in leaf, wood, and root concentrations throughout an annual cycle. Seasonal variation in radiocesium content of aquatic vegetation was observed by Gladden (1979).

Shure and Gottschalk (1975) reported strong seasonality effects on radiocesium concentrations of physical and biotic components within a Coastal Plain stream. Temporal variation of radiocesium concentration in wild birds was reported by Levy, et al. (1975). Temporal changes in cesium-137 concentration in fish have been observed (Smedile and Queirazza 1975) and were related to physiological activity and nutrient conditions of the water. These examples of temporal patterns of radionuclide concentration in ecosystems along with many others in the literature serve to establish the fact that organisms will contain varying concentrations of biologically active radionuclides over an annual cycle and that physical components of the ecosystem will have changing concentrations which may be cyclic or directional over time.

#### DISCUSSION

The comparison of single species to ecosystem assessment of radionuclear ecotoxigants shows that the single species approach is not sufficient to simulate the actual impacts and transport of these ecotoxigants as observed in functioning ecosystems. The single species or microcosm assessments fail simply because they cannot adequately simulate the enormous complexity and variety of physical, chemical, and biological processes of ecosystems at the large scale at which ecosystems function.

The use of ecosystem methods to develop our knowledge of radionuclear ecotoxigants produced results which differ considerably from the knowledge we would have gained from single species tests. These results include, but are not limited to, the following:

1. The environmental behavior of radionuclides varies both within and across physiographic provinces.
2. Dispersed radionuclear ecotoxigants may be reconcentrated in a

food chain with bioamplification at higher trophic levels and in larger herbivores and carnivores.

3. Organisms such as lichens and fungi which concentrate radionuclear ecotoxigants to comparatively high levels are highly significant in food chain bioamplification and recycling of radionuclear ecotoxigants.
4. Temporal variation of radionuclear concentrations in organisms, either as seasonal phenomena or as a directional change over time, is characteristic of organisms in ecosystems.
5. Naturally occurring physical processes may transport radionuclear ecotoxigants between ecosystems producing accumulation and reconcentration of these ecotoxigants in the terminal ecosystem.
6. Most organisms exhibit differential life-stage radiosensitivities such that over the life history of an organism a constant environmental radiation dose could have variable effects ranging from no impact to lethal impact.
7. Single-species-based predictions of mortality from ionizing radiation are not necessarily realized when a "lethal" dose is delivered to a species in an ecosystem. Conversely, doses determined to be "sub-lethal" through single species tests may be lethal to a species when the "sub-lethal" dose is delivered to the species living in an ecosystem.
8. The impacts of ionizing radiation delivered to organisms in an ecosystem are greater than the simple sum of the impacts on each individual. These system-based impacts include changes in species diversity, productivity, nutrient cycling, and possible long-term exclusion of some species. This greater damage to the ecological

system results from species-species interactions and dependencies and species-environment interactions.

The conclusion is that single species or microcosm tests have a useful, but limited, role in defining radionuclear impacts on organisms. Some of the uses of single species testing of radionuclear ecotoxicants are to explain phenomena observed at the ecosystem level, to provide preliminary trials prior to large scale ecosystem testing, to study in greater, isolated detail, specific mechanisms identified from ecosystem testing, to develop a baseline of potential impact to life, to develop predictive relationships between dose and response, and to analyze damage mechanisms. However, in spite of the uses and attractiveness of single species testing, it does not provide the quality of data sufficient for realistic assessment of radionuclear impacts. In comparison, ecosystem testing provides comprehensive and conclusive knowledge of radionuclear ecotoxicant impacts which allows analysis of short-term and long-term impacts, provides data whose transferability to other systems and regions can be evaluated, allows extrapolation with maximum confidence, and allows explicit comparison of the severity of impact from a variety of ecotoxicants. In this analysis, single species testing is seen as a highly useful, but supplemental, method to ecosystem testing. Under the constraint that only one method could be used, ecosystem testing would be the preferred method for evaluating the impact of radionuclear ecotoxicants.

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LITERATURE CITED

- Auerbach, S. I., D. A. Crossley and A. F. Shinn. 1968. Postattack insect problems. pp. 137-142. In: Proceedings of the Symposium on Post Attack Recovery from Nuclear War, (anonymous). National Academy of Science, National Academy of Engineering, National Research Council, Washington, D. C. vii, 434 pp.
- Bond, V. P. 1969. Radiation mortality in different mammalian species: pp 5-19. In: Comparative Cellular and Species Radiosensitivity; V. P. Bond and T. Sugahara (eds.) Igaka Shoin, Tokyo.
- Chappell, H. G. 1963. The effect of ionizing radiation on Smilax with special reference to the protection afforded by their production of underground vegetative structures. pp. 289-294. In: Radioecology, V. Schultz and A. Klement, Jr. (eds.). Reinhold Publishing Co., NY.
- Cotter, D. J. and J. T. McGinnis. 1965. Recovery of hardwood stands 3-5 years following acute radiation. Health Phys. 11:1663-1673.
- Dahlman, R. C., C. W. Francis and T. Tamura. 1975. Radiocesium cycling in vegetation and soil. pp. 462-481. In: Mineral Cycling in Southeastern Ecosystems, F. G. Howell, J. B. Gentry and M. H. Smith (eds.). CONF-740513. NTIS, Springfield, VA.
- Fraley, L. Jr., and F. W. Whicker. 1973. Response of shortgrass plains vegetation to gamma radiation--I. Chronic Irradiation. In: Rad. Bot. 13:331-341.
- Frank, Marilyn L. and B. G. Blaylock. 1971. Effects of acute ionizing radiation on carp (Cyprinus carpio L.) embryos. ORNL-TM-3346. 48 pp.
- Gamble, John F. 1971. A proposed mechanism for the recycling of radiocesium in Florida soil systems. pp. 133-139. In: Radionuclides in Ecosystems, D. J. Nelson (ed.). CONF-710501-P1. NTIS, Springfield, VA

- Garten, C. T., L. A. Briese, R. R. Scharitz and J. B. Gentry. 1975. Radiocesium levels in vegetation colonizing a contaminated floodplain, pp. 489-497. In: Mineral Cycling in Southeastern Ecosystems, F. G. Howell, J. B. Gentry and M. H. Smith (eds.) CONF-740513, NTIS, Springfield, VA.
- Garten, C. T., L. A. Briese, R. R. Sharitz, and J. B. Gentry. 1975. Seasonal variation in radiocesium concentrations in three tree species. pp. 498-508. In: Mineral Cycling in Southeastern Ecosystems, F. G. Howell, J. B. Gentry and M. H. Smith (eds.) CONF-740513. NTIS, Springfield VA.
- Gladden, John B. 1979. Radiocesium exchange among components of a Coastal Plain bottomland stream ecosystem. Ph.D. Dissertation. Emory University, Atlanta, GA. 191 p.
- Gunkel, J. E. and A. H. Sparrow. 1961. Ionizing radiations: biochemical, physiological and morphological aspects of their effects on plants. pp. 555-611. In: W. Ruhland (ed.), Encyclopedia of Plant Physiology. Vol. XVI. Springer Verlag, Berlin.
- Hanson, W. C. and H. E. Palmer. 1965. Seasonal cycle of cesium-137 in some Alaskan natives and animals. Health Phys. 11:1401-1406.
- Hanson, W. C. 1967. Radioecological concentration processes characterizing arctic ecosystems. pp. 191-193. In: Radioecological Concentration Processes, B. Aberg and F. P. Hungate (eds.) Pergamon Press, Oxford.
- Hanson, W. C. and L. L. Eberhardt. 1971. Cycling and compartmentalizing of radionuclides in northern Alaskan lichen communities. In: Radionuclides in Ecosystems, D. J. Nelson (ed.) CONF-710501-P1. NTIS, Springfield, VA.
- Hay, J. D. and H. L. Ragsdale. 1978. Patterns of cesium-137 distribution across two disparate floodplains. pp. 462-478. In: Environmental Chemistry and Cycling Processes, D. C. Adriano and I. L. Brisbane, Jr. (eds.). CONF-760429. NTIS, Springfield, VA.

- Heaslip, Margaret B. 1959. Effects of seed irradiation on germination and seedling growth of certain deciduous trees. In: Ecology. 40:383-388.
- Holleman, D. F., and J. R. Luick. 1975. Relationship between potassium-intake and radiocesium retention in the reindeer. pp. 557-563. In: Mineral cycling in southeastern ecosystems, F. G. Howell, J. B. Gentry and M. H. Smith (eds.). CONF-740513. NTIS, Springfield, VA.
- Jenkins, J. H., J. R. Monroe and F. B. Golley. 1967. Comparison of fallout cesium-137 accumulation and excretion in certain southeastern mammals. pp. 623-626. In: Symposium on Radioecology, D. J. Nelson and F. C. Evans (eds.). CONF-670503. NTIS, Springfield, VA.
- Jenkins, J. H. and T. T. Fendley, 1968. The extent of contamination, detection, and health significance of high accumulation of radioactivity in southeastern game populations. Proc. S. E. Association Game and Fish Comm. 22:89-95.
- Jenkins, James H. and Timothy T. Fendley. 1971. Radionuclide biomagnification in Coastal Plain deer. pp. 116-122. In: Radionuclides in Ecosystems, D. J. Nelson (ed.). CONF-701501-P1. NTIS, Sprigfield, VA.
- Klement, A. W., Jr. 1965. Radioactive fallout phenomena and mechanisms. Health Phys. 11:1265-1274.
- Larson, Clarence E. 1971. Not in Retrospect. pp. 13-16. In: Radionuclides in Ecosystems, D. J. Nelson (ed.). CONF-710501-P1. NTIS, Springfield, VA.
- Larson, Kermit H. 1963. Continental close-in fallout: its history, measurement and characteristics. pp. 19-25. In: Radioecology, V. Schultz and A. Klement, Jr. (eds.). Reinhold Pub. Co., NY.

- Levy, C. K., K. A. Youngstrom and C. J. Muletskos. 1975. Whole body gamma-spectroscopic assessment of environmental radionuclides in recapturable wild birds. pp. 113-122. In: Radioecology and Energy Resources, Colbert E. Cushing, Jr. (ed.) Dowden, Hutchinson and Ross, Inc, Stroudsburg, PN.
- McCormick, Frank. 1963. Changes in a herbaceous community during a three-year period following exposure to ionizing radiation gradients. pp. 271-276. In: Radioecology, V. Schultz and A. W. Klement, Jr. (eds.) Reinhold Pub. Co., NY.
- McCormick, J. Frank and Robert E. McJunkin. 1965. Interaction of gamma radiation and other environmental stresses upon pine seeds and seedlings. Health Phys. 11:1643-1652.
- Menhenick, Edward F. and D. A. Crossley, Jr. 1968. A comparison of radiation profiles of Acheta domesticus and Tenebrio molitor. Annals of the Entomological Society of America. 61:1359-1365.
- Menhenick, Edward F. and D. A. Crossley, Jr. 1969. Radiation sensitivity of twelve species of arthropods. Annals of the Entomological Society of America. 62:711-717.
- McCormick, J. Frank. 1967. Effects of ionizing radiation on a pine forest. pp. 78-87. In: Symposium on Radioecology, D. J. Nelson and F. C. Evans (eds.). CONF-670503. NTIS, Springfield, VA.
- Miller, Carl F. 1971. Properties of fallout important to agriculture. pp. 81-106. In: Survival of Food Crops and Livestock in the Event of Nuclear War, D. W. Benson and A. H. Sparrow (eds.). CONF-700909. NTIS, Springfield, VA.
- Murphy, Peter G. and J. Frank McCormick. 1971. Ecological effects of acute beta irradiation from simulated fallout particles on a natural plant community. pp. 454-481. In Survival of food crops and livestock in event of nuclear war, D. W. Benson and A. H. Sparrow (eds.). NTIS. Springfield, VA.

- Patten, B. C. and M. Witkamp. 1967. System analysis of cesium-137 in terrestrial microcosms. *Ecology* 48:813-824.
- Platt, R. B. 1963. Ecological effects of ionizing radiation on organisms, communities and ecosystems. pp. 243-255. In: *Radioecology*, V. Schultz, A. W. Klement, Jr. (eds.). Reinhold Pub. Co., NY.
- Platt, R. B. 1965. Radiation effects on plant populations and communities: research status and potential. *Health Phys.* 11:1601-1606.
- Radioactive effluents from nuclear fuel reprocessing plants. 1978. Comm. of European Nations. Karlsruhe, Germany. 762 p.
- Ragsdale, H. L., J. P. Witherspoon and D. J. Nelson. 1968. The effects of biotic complexity and fast neutron radiation on cesium-137 and cobalt-60 kinetics in aquatic microcosms. 174 p. ORNL-4318. ORNL, Oak Ridge, TN.
- Ragsdale, H. L. and D. J. Shure. 1973. Floodplain transfer and accumulation of cesium-137 from a reactor effluent stream, pp. 243-253. In: *Environmental Behavior of Radionuclides Released in the Nuclear Industry*. Inter-Atomic Energy Agency, Vienna. IAEA-SM-172-33.
- Ragsdale, H. L. and W. A. Rhoads. 1974. Four-year post exposure assay of vegetation surrounding Project Pinstripe: Demonstration of the utility of delayed damage appraisals. *Rad. Bot.* 4:229-236.
- Rhoads, W. A., H. L. Ragsdale, R. B. Platt and E. M. Romney. 1971. Radiation doses to vegetation from close-in fallout at Project Schooner. pp. 352-369. In: *Survival of Food Crops and Livestock in the Event of Nuclear War*, D. W. Benson and A. H. Sparrow (eds.). COND-700909. NTIS, Springfield, VA.
- Ritchie, J. C., J. R. McHenry and G. D. Bubenzer. 1980. Redistribution of fallout cesium-137 in Brunner Creek Watershed in Wisconsin. Prepublication manuscript. J. C. Ritchie, Staff Scientist, USDA, Sea-Agricultural Research, Beltsville, MD.

- Sharitz, R. R., S. L. Scott, J. E. Pinder, III and S. K. Woods. 1975. Uptake of radiocesium from contaminated floodplain sedimentsd by herbaceous plants. Health Phys. 28:23-28.
- Shure, D. J. and M. R. Gottschalk. 1975. Cesium-137 in a reactor effluent stream. pp. 234-241. Radioecology and Energy Resources, Colbert E. Cushing, Jr. (ed.). Dowden, Hutchinson and Ross, Inc. Stroudsburgh, PN.
- Shure, D. J. and M. R. Gottschalk. 1978. Radiocesium transfer through aerial pathways in a South Carolina Floodplain Forest. pp. 709-724. In: Environmental Chemistry and Cycling Processes, D. C. Adriano and I. L. Brisbane, Jr. (eds.) CONF-760429. NTIS, Springfield, VA.
- Smedile, Elio and Guilio Queirazza. 1975. Uptake of cobalt-60 and cesium-137 in different components in a river exosystem connected with discharges of a nuclear power station. pp. 1-8. In: Radioecology and Energy Resources, Colbert E. Cushing, Jr. (ed.). Dowden, Hutchinson and Rose, Inc. Stroudsburg, PA.
- Sparrow, A. H. 1963. Comparisons of the tolerances of high plants to acute and chronic ionizing radiation. Japan J. Gen. 40:12-37.
- Sparrow, A. H., R. C. Sparrow, K. H. Thomson and L. A. Schairer. 1965. The use of nuclear and chromosomal variables in determing and predicting radiosensitivities. Rad. Bot. (suppl) 5:101-132.
- Sparrow, A. H. 1966. Research uses of the gamma field and related radiation research facilities at Brookhaven National Laboratory. Rad. Bot. 6:377-405.
- Sparrow, A. H., A. F. Rogers and S. S. Schwemmer. 1968. Radiosenstivity studies with woods plants. I. Acute gamma irradiation survival data for 28 species and predictions for 190 species. Rad. Bot. 8:149-166.
- Tamura, T. and D. G. Jacobs. 1961. Structural implications in cesium sorption. Health Physics. 2:391-398.

- Waste Isolation Safety Assessment Program Technical Progress Report for FY-1978. 1979. Pacific Northwest Laboratory, PNL-2874. Richland, Washington.
- Wegener, K. H. 1966. Radioaktivitat und veterinarmedizen. Paul Parey, Berlin.
- Witherspoon, J. P. 1965. Radiation damage to forest surrounding an unshielded fast reactor, Health Phys. 11:1637-1642.
- Witherspoon, J. P. 1967. Radiosensitivity of forest trees to acute fast neutron radiation. pp. 120-126. In: Symposium on Radioecology, D. J. Nelson and F. C. Evans (eds.) CONF-670503. NTIS, Springfield, VA.
- Witherspoon, J. P. 1968. Effects of internal cesium-137 radiation on seeds of Liriodendron tulipifera. Rad. Bot. 8:45-48.
- Witkamp, M. and M. L. Frank. 1967. Cesium-137 kinetics in terrestrial microcosms. pp. 635-643. In: Symposium on Radioecology. D. J. Nelson and F. C. Evans, Jr. (eds.). CONF-670503. NTIS, Springfield, VA.
- Witkamp, Martin and V. A. Merchant. 1971. Effects of light, temperature and soil fertility on distribution of manganese-54 and cesium-137 in producer-consumer microcosms. pp. 204-208. In: Radionuclides in Ecosystems, D. J. Nelson (ed.). CONF-710501-P1. NTIS, Springfield, VA.
- Woodwell, G. M. 1962. Effects of ionizing radiation on terrestrial ecosystems. Science. 138:572-577.
- Woodwell, G. M. and A. L. Rebeck. 1967. Effects of chronic gamma irradiation on the structure and diversity of an oak-pine forest. Ecolog. Monogr. 37:53-69.

- Woodwell, G. M. and B. R. Holt. 1971. Effect of nuclear war on the structure and function of natural communities: an appraisal based on experiments with gamma radiation. pp. 482-491. In: Survival of Food Crops and Livestock in the Event of Nuclear War, D. W. Benson and A. H. Sparrow (eds.). CONF-700909. NRIS, Springfield, VA.
- Zavitkovski, J. and T. D. Rudolph. 1971. Predicted effects of chronic gamma irradiation on northern forest communities. pp. 1007-1014. In: Radionuclides in Ecosystems, D. J. Nelson (ed.). CONF-710501-P2. NTIS, Springfield, VA.



CLASSES OF ECOTOXICOLOGICAL TESTS:  
THEIR ADVANTAGES AND DISADVANTAGES FOR REGULATION

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Abstract

Three classes of ecotoxicological tests, single-species, microcosm, and field tests, are discussed and evaluated for their usefulness in toxic substance regulation. Criteria for evaluation are ability to predict ecosystem-level effects, replicability, standardizability, speed, simplicity, and cost. The major conclusion is that microcosms of intermediate complexity and small field plots are presently the most useful tools for ecotoxicological testing and hazard evaluation.

## INTRODUCTION

EPA's charge to the National Academy Ecotoxicology Review Committee is to evaluate and recommend tests and methods for determining whether chemicals have toxic effects to ecosystems above the level of individual species (NRC, 1979). There are three broad classes of tests in ecotoxicology that can be considered: single-species tests, tests conducted on laboratory and field microcosms, and field tests. For purposes of this review, we do not consider mathematical modeling to constitute a test method. The present best use of mathematical models in ecotoxicology is in conjunction with the three listed test methods, as a means of searching for correlations among data, of extrapolating laboratory results to a natural context, and of identifying sensible experiments and test procedures. In the pages that follow each of the three classes of tests will be described in more detail, their respective advantages and limitations discussed, and evaluations made of their potential usefulness for ecotoxicological regulation.

In order for ecotoxicological tests to be of value for regulation, they must meet several criteria (Government, 1979). First and foremost they should be predictive. Tests should indicate correctly which ecosystems are at risk, and within those systems for what specific components, at what chemical concentrations, and under what environmental conditions there should be concern. Second, tests must be replicable (i.e., reproducible) and standardized. Replicability ensures that test results can be statistically supported. Standardization permits widespread accessibility to test procedures and materials for individuals and firms interested in chemical testing. Furthermore, standardized tests ease EPA's task of result analysis and subsequent risk assessment. Third, tests should be relatively rapid, simple, and as inexpensive as practicable. These characteristics are desirable to forestall excessive delays in

hazard evaluation, to permit efficient use of limited technical and physical resources available for testing, and to prevent stifling of innovation for substances whose production volumes may not be profitable enough to support an expensive batch of premanufacture testing. The classes of ecotoxicological tests described below will be judged with respect to these criteria.

#### SINGLE-SPECIES TESTS

Tests conducted on individual species comprise an important class of tests to determine the toxic effects of chemicals. In these tests, generally performed under controlled laboratory conditions, groups of test organisms are exposed to a range of chemical concentrations and responses observed at each concentration are recorded. Dose-response curves are then constructed from these data. The usefulness of such curves is that any expected environmental concentration of the chemical can be selected and the likely response of the test organism determined from the graph.

There are a number of virtues to such tests. Results are often relatively easy to see. This is particularly true when mortality is being observed. For plant and microbe populations, death is sometimes harder to measure. In these cases some other function, such as growth or respiration, usually can be monitored. When compared to control populations, toxic effects can then be inferred. A number of standardized tests are available, allowing for good replicability, widespread accessibility, and lower cost (Draggan, 1978). As a rule these tests are relatively rapid, and therefore not particularly consumptive of facilities and personnel. An exception is chronic toxicity testing for higher level species (e.g., fish), which can take up to several years. A final advantage to single-species tests is that some guidelines for their use already exist. The EPA has issued recommendations for what tests could be of value in its process of pre-manufacture assessment of risk under the Toxic Substances Control Act (EPA, 1979).

These advantages must be weighed against a number of important drawbacks. Tests conducted on single species are limited in what they can reveal about interspecies interactions and about the relationships of a biological community with its environment. A predator-prey example by Taub (1976) illustrates this point. Suppose that a prey species is involved in a single-species test, and that a toxic chemical reduces the growth rate of the prey population. If both the birth and the death rates of the population are lowered (the latter perhaps by lowering the rate of cannibalism), then the stock (i.e., number) of prey may remain the same but the flow of biomass through the population would be reduced. A predator population, which survives on the flow of biomass, will lose a source of food. If that source is crucial to the survival of the predator, the predator population could become extinct--and no change would have been observed in the size of the prey population. It is true that by measuring the flow rather than the stock of the prey population, the effect on the predator population could have been anticipated; in general, however, not enough characteristics will be monitored in a single-species tests to adduce all indirect effects. A more convincing example would be a chemical that alters succession patterns in a biological community. Such effects could hardly be inferred simply by observing one species. The general point being made is that ecosystems are complex, interlocking sets of components, some of whose properties arise not from the components themselves, but from the specific set of interactions within the system. Therefore, it is not possible to characterize the system and its likely responses to perturbations solely from a knowledge of the component parts (Heath, 1979).

Certain exceptions to this statement can be made. When biochemical pathways bottleneck or when an important role in ecosystem functioning is localized in one species, genus, or trophic level, then that species or level can be used as an

indicator of biological integrity. For example, in the nitrogen cycle the process of nitrification is funneled through the bacterial genus Nitrobacter. Should a substance prove toxic to Nitrobacter, it can be expected to disrupt the nitrogen cycles of any ecosystems exposed to it. In a different case, Kaesler and Cairns (1972) have evidently shown that diatoms are "useful for estimating the response of the entire aquatic community" (cited in Cairns et al., 1976). Indicators of ecological integrity may turn out to be some of the most useful and important tools for screening out toxic substances from the commercial inventory. At the present time, however, they remain a subject of ongoing research and are not generally available.

A second problem with single-species tests concerns the question of their realism. This issue, which will also arise in the discussion of microcosms, can be conveniently discussed by introducing the terms "ecological realism" and "pollution realism" (Blanck et al., 1978). Ecological realism indicates how well the test system accounts for important characteristics of the ecosystem part of a polluted environment. Organisms in their natural setting are constrained in numerous ways not found in the laboratory. Competition for nutrients, space, light; pressures from predators and parasites; and a host of other factors operate simultaneously in natural environments to prevent a species from reaching its maximum size, range, and relative abundance. A species isolated and in the generally optimal conditions of a laboratory test cannot be expected to respond identically to the way it would in its constrained natural setting. In a similar vein, pollution realism indicates how well the test system accounts for the state of pollution in a polluted environment. A pollutant is not generally found in isolation: there are usually other exogenous substances in the environment that potentially can enhance or diminish a substance's effect. In addition, both the biotic and abiotic parts of ecosystems can alter the

concentration, chemical form, and dose-rate by which a given species is exposed to a chemical. A laboratory system cannot be expected to mimic the exposure conditions, and therefore elicit the typical response, that would be observed in the wild. For these reasons systems exhibiting more realism than do single-species tests are being developed.

#### MICROCOSM TESTS

In order to overcome some of the difficulties of single-species tests, more complicated test systems are being developed. Such systems, using simulated ecosystems, are called microcosms, microecosystems, or model ecosystems. Microcosms span a continuum of complexity from simple two-species systems at one extreme to segments of natural ecosystems isolated for experimentation at the other. They are typically of small size so they can be constructed in the laboratory or in reasonably constrained field situations. Microcosms are of two general types: those constructed from scratch, the compositions of which are known exactly (gnotobiotic systems), and those whose bases are portions of natural ecosystems to which some components have been added or removed. Each of these types has respective advantages and disadvantages: the former, more accurately known composition; the latter, more realism. But both types exhibit a level of complexity and realism surpassing single-species systems (e.g., interactions are present among biota as well as between the biota and their surroundings), and at the same time they provide a certain amount of simplification compared to natural systems to ease the study of key features.

The potential advantages of using microcosms as test systems are considerable (Draggan, 1976; Harte et al., 1979). Their compactness and common environments permit both replication and standardization. The chemical composition of the medium and the trophic structure can be manipulated easily so that analogs of

qualitatively different ecosystems can be created. The uniform conditions that can be created facilitate the comparison of different substances. The lack of complicated spatial heterogeneity allows for more complete definition of physical, chemical, and biological characteristics. Causal relationships are more easily inferred than in natural systems because of the absence of complicating environmental variability. Perturbations of different physical, chemical, and biological variables can be carried out with little effort and expense. Potentially dangerous test substances and radiotracers can be administered without contamination of the general environment. (Although contaminated microcosms must be disposed of eventually, this could occur in more carefully selected depositories, e.g., in radioactive waste burial sites, than the general release of chemicals would entail.) Rapid evaluation is often possible with microcosms. And perhaps most importantly, ecosystem-level effects can often be observed.

Against these potential advantages several limitations must be assessed. Again the twin questions of ecological and pollution realism arise. Microcosms are intentional simplifications of real systems. As a result, responses observed in microcosms must be extrapolated to expected responses of actual ecosystems. For very simple microcosms there is the obvious problem that significant aspects of the natural systems are absent. These components could exert crucial influences in the responses of the natural ecosystems to stress. As microcosms become progressively more complex and include progressively more of the biotic and abiotic components of their natural counterparts, this problem becomes smaller. But there always remains residual differences between even the most ecologically complex model systems and their natural counterparts.

The small size of most microcosms introduces unavoidable problems of scaling that further reduces their ecological realism. Some of the problems of scale are illustrated in aquatic microcosms (Dudzik, 1979; Harte *et al.*, 1979;

Jassby et al., 1977a, 1977b; Whittaker, 1961). The shallow depths of most aquatic microcosms allow benthic compartments to exert unrealistically large effects on nutrient fluxes and decomposition activities. Shallow depths also distort zooplankton vertical migration patterns and the losses of phytoplankton from the water column due to sinking. The inclusion of macrofauna such as fish, snails, and larger crustacea, while desirable in order to add higher trophic levels to the systems, can overwhelm nutrient cycles in small microcosms. The high surface-to-volume ratios of most microcosms permit side and bottom effects (e.g., periphyton growth) to exert disproportionately large influences compared to natural systems. Realistic conditions of water mixing and thermal stratification are difficult to produce, and in the latter case, perhaps undesirable as well, because the small hypolimnion that could be created would be quite unrepresentative of natural lakes. Small systems may also suffer problems of small numbers: dramatic fluctuations in percentage population size may appear because the absolute sizes of such populations are low. These fluctuations in turn may mask important but subtle effects that are taking place.

In the area of pollution realism much the same can be said for microcosms as was said for single-species tests. Toxic substances present together in natural systems can interact to enhance or diminish each other's effects. But substances are generally tested singly in microcosm research. Both the biological and abiological components of natural ecosystems influence the exact exposure patterns of organisms to toxic chemicals. The rendering of inorganic mercury to the more hazardous methylmercury by certain micro-organisms is a familiar example. In very simple model ecosystems many of these influences will be absent. For more complex and ecologically realistic microcosms, this is less of a problem. But if more complex model systems are used, some of the advantages of microcosms--ease of construction and manipulation; better understanding of

interactions taking place; etc.--disappear.

Lastly, the potential advantage associated with the replicability of microcosms is not fully established and is the subject of ongoing research (Isensee, 1976; Crow and Taub, 1979). In fact, microcosm research is relatively new and virtually all aspects of microcosm use are currently under study. Nevertheless the interest in microcosms for both basic and applied ecological research is substantial as is demonstrated by the number of symposia held on them in recent years (see, for example, the November 1976 and January 1979 special issue of the International Journal of Environmental Studies and Giesy, 1979).

#### FIELD TESTS

Test conducted on naturally occurring ecosystems in the field are the most realistic of all tests. Whether the test is on a small scale, as in a pond or stream, or on a large scale, as in a forest or watershed, the unconstrained nature of the system is the defining characteristic. There are few extrapolations to be made in tests of this sort: the systems examined are selected because they are examples of systems that would normally be exposed to the chemical under test. Subtle effects and interactions not present in simplified systems can be expected to occur in the field (NAS, 1975). Species at the tops of food chains may be especially sensitive, for example. Yet these species are difficult to incorporate into simplified systems, as discussed above in connection with aquatic microcosms. Biomagnification processes could therefore be difficult to detect except in the field. Similarly the effects of chemicals whose biological breakdown products are themselves toxic would be difficult to observe in the laboratory. The pollution situation in the field, insofar as other contaminants is concerned, is likely to resemble that found when the chemical is actually

used (Cairns and Dickson, 1978). With regard to both ecological and pollution realism, then, field tests are the most desirable tests. Field tests may also be necessary to validate results obtained with laboratory systems, at least in the latter's earlier stages of development, to ensure that the microcosm results are robust.

Despite these important advantages, field tests are not necessarily indicated (Cook, 1971; NAS, 1975; Draggan, 1976; Lighthart and Bond, 1976; Heath, 1979). Field tests are difficult to replicate because environmental conditions are neither uniform spatially nor temporally. The Experimental Lakes Area of Northwestern Ontario, Canada, is a rare, perhaps unique, exception in that hundreds of virtually identical small lakes are available for research and there can be a high level of replicability (Johnson and Vallentyne, 1971; and all articles in that issue). In general, however, such conditions of natural replicability will not be found. Natural background variability makes field tests difficult to interpret unequivocally. That is, test results can always be ascribed to variable factors other than to the chemical under study. Spatial heterogeneity of ecosystems can lead to the requirement that large and numerous sites be investigated to encompass the wide range of ecosystem exposures a chemical may produce. This in turn could lead to widespread contamination. Temporal environmental variations simply may be impossible to examine in the field. For instance, a substance may be especially detrimental only at times of other environmental stress, for example, during a prolonged drought. Such extremes in environmental conditions are usually not conveniently available during field testing. Even the advantage of pollution realism mentioned above is not so secure when examined closely. A chemical to be released into a polluted river, for example, would not ordinarily be field tested in that same river. Therefore the chance that the exact pollution condition will be created

in the test is small. In addition, field tests can be lengthy, expensive, and consumptive of personnel, often tying up resources out of proportion to the benefits derived. And perhaps most importantly, the natural environment is limited yet the list of chemicals to be tested seems endless. There are simply not enough natural systems to go around to field test every prospective new chemical (or existing ones for that matter) without defeating the whole purpose of the screening procedure.

#### EVALUATION OF THE TEST TYPES, SUMMARY, AND CONCLUSIONS

To decide which types of tests are most useful for ecotoxicological regulation, they must be compared on the bases described above: ability to predict ecosystem-level effects, replicability, standardizability, rapidity, ease, and cost (see P. 1). There are hazards in making such comparisons, however, because some criteria are more important than others yet it is difficult to give an explicit weight to each criterion. For example, a test that is fast, reproducible, and inexpensive--a single-species test, perhaps--may well be considered to be of less value than a slower, more expensive, less replicable test that is, however, better at predicting ecosystem-level effects--a microcosm test, perhaps. In principle each criterion could be given a weight representing its importance to the goal of effective ecotoxicological regulation. This could be done for instance by polling competent specialists in the field and averaging their individual rankings of criterion importance. Yet this procedure is cumbersome and perhaps no more effective than making a specific judgment based on the particulars of the chemical in question and the various candidate tests available at the time a decision must be made. For example, a chemical with a potentially widespread exposure and suspected hazard would indicate a need for more searching tests than would a chemical of limited release or of no suspected hazard. These judgments would be difficult to formalize into an explicit

weighting procedure; perhaps some sort of guidance would be needed from EPA to indicate to chemical manufacturers which types of tests are most useful in which situations. This problem may be resolved when a decision is finally made on the structure of the testing procedure (tier testing versus battery testing, etc.). However, such a decision does not eliminate all major problems (EPA, 1979; Conservation Foundation, 1978).

Nevertheless, with due regard for the above problem, it does seem possible to gain an overview of which test types are likely to be most valuable for regulation, based on the major features presented in the preceding sections. Single-species tests rank high in a number of areas. They are (or can be) standardized, replicable, relatively fast, inexpensive, and simple. Their major flaw, an inability to successfully predict ecosystem-level effects, is however a crucial one in this particular area of toxic substances regulation. Until effective indicators of ecological effects have been discovered, that is, single species whose intact functioning indicates ecological integrity, it would seem that single-species tests are of limited value for ecotoxicological regulation.

Microcosm tests can be simple or complex. The simpler ones tend not to model very well important ecosystem functions; the complex ones are difficult to conduct, to replicate, and to incorporate realistic ecological characteristics (see the discussion above on the problems of aquatic microcosms). At this stage of development, microcosms of intermediate complexity, which include some but not all ecosystem features of interest, appear to hold the most promise for being reliable test systems. Because this conclusion--that microcosms of intermediate complexity are the most useful ones--is central, it is worthwhile elaborating a little on what is meant by intermediate complexity.

Basically there are three degrees of freedom along which complexity can be measured: biological, physical, and chemical. Biological complexity can be

measured in terms of number of species, their population characteristics, and the interrelationships among them. For example, a very simple system would include one or several species, representing one or perhaps two trophic levels. A very complex system would contain many species, probably complex age structures, representing many trophic levels. Intermediate in complexity would be a system containing a large number of species but probably not those of the highest trophic levels. In an accompanying paper Harte describes a whole-water initiation strategy in which lakewater containing all plankton species but no macrofauna is used to stock microcosms. Other examples can be conceived. A physically complex system would try to incorporate natural patterns of temperature, light, moisture, turbulence, etc. A physically simple system would select constant values for most of these variables. And a system of intermediate complexity would try to vary at least some of these factors in at least semi-realistic ways (e.g., a 12 hour light: 12 hour dark photoperiod). In a similar way, chemically complex systems would try to model natural sources of many nutrients and chemicals as well as potential toxicants, including their natural time variation (e.g., a pulse of nutrients to represent spring thaw in lakes). Simple systems would hold chemical values constant or allow them to increase or decrease from an initial load. Systems intermediate in complexity would have their chemical concentrations determined by the organisms within and by the trophic relations among those organisms. Experts in different areas of microcosm research can fill in the relevant details from this sketchy description.

A compromise between the goals of realism and of practicality is admittedly being made in drawing this conclusion about microcosms of intermediate complexity. Nevertheless, the state of microcosm research seems to point unavoidably in this direction. The other advantages of microcosm tests, their standardizability, replicability, and ease of manipulation, indicate that microcosm tests are likely

to be of high value in toxic substances regulation. Field tests can be extremely realistic. However, environmental conditions are difficult or impossible to control, making replicability problematic and the modeling of qualitatively different environmental conditions virtually impossible. In addition, field tests would be difficult to standardize and make accessible to a broad spectrum of potential users. There exists, finally, the potential for substantial environmental contamination if even small test areas are used for a large number of substances.

In conclusion, the most likely useful types of tests for ecotoxicological regulation, the ones most deserving of attention and research effort, are microcosms of intermediate complexity and small field plots. This happens also to be the conclusion of a National Academy of Sciences study made five years ago (NAS, 1975).

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REFERENCES

- Blanck, H., G. Dave, and K. Gustafsson. 1978. An Annotated Literature Survey of Methods for Determination of Effects and Fate of Pollutants in Aquatic Environments. Report from the National Swedish Environment Protection Board.
- Cairns, J. Jr., K. L. Dickson, J. P. Slocumb, S. P. Almeida, and J. K. T. Eu. 1976. Automated pollution monitoring with microcosms. Intern. J. Environmental Studies 10: 43-49.
- Cairns, J. Jr., and K. L. Dickson. 1978. Field and laboratory protocols for evaluating the effects of chemical substances on aquatic life. J. Testing and Evaluation 6 (2): 81-90.
- Cook, G. D. 1971. Aquatic laboratory microcosms and communities. pp. 48-85 in J. Cairns, Jr. (ed.), The Structure and Function of Freshwater Microbial Communities (Virginia Polytechnic Institute and State University).
- Conservation Foundation, 1978. Approaches for Developing Testing Guidelines Under the Toxic Substances Control Act.
- Crow, M. E. and F. B. Taub. 1979. Designing a microcosm bioassay to detect ecosystem level effects. Intern. J. Environmental Studies 13: 141-147.
- Draggan, S. 1976. The microcosm as a tool for estimation of environmental transport of toxic materials. Intern. J. Environmental Studies 10: 65-70.
- Draggan, S. 1978. TSCA: The U. S. attempt to control toxic chemicals in the environment. Ambio 7 (5/6): 260-262.
- Dudzik, M., J. Harte, A. Jassby, E. Lapan, D. Levy, and J. Rees. 1979. Some considerations in the design of aquatic microcosms for plankton studies. Intern. J. Environmental Studies 13: 125-130.

- Environmental Protection Agency. 1979. Toxic Substances Control: Discussion of premanufacture testing policy and technical issues; request for comment. Federal Register 44 (53): 16240-16292 (16 March 1979).
- Giesy, J. (ed.). 1979. Proceedings from the Symposium on Microcosms in Ecological Research (to be published).
- Government's Statement of Work. 1979. Annex II. EQ9AC008.
- Harte, J., D. Levy, J. Rees, and E. Saegebarth. 1979. Making microcosms an effective assessment tool, in J. Giesy (ed.), Proceedings from the Symposium on Microcosms in Ecological Research (to be published).
- Heath, R. T. 1979. Holistic study of an aquatic microcosm: theoretic and practical implications. Intern. J. Environ. Studies 13: 87-93.
- Isensee, A. R. 1976. Variability of aquatic model ecosystem-derived data. Intern. J. Environmental Studies 10: 35-41.
- Jassby, A., M. Dudzik, J. Rees, E. Lapan, D. Levy, and J. Harte, 1977a. Production cycles in aquatic microcosms. Environmental Protection Agency. Office of Research and Development Research Report EPA-600/7-77-097. National Technical Information Service, Springfield, VA.
- Jassby, A., J. Rees, M. Dudzik, D. Levy, E. Lapan, and J. Harte, 1977b. Trophic structure modifications by planktivorous fish in aquatic microcosms. Environmental Protection Agency. Office of Research and Development Research Report EPA-600/7-77-096. National Technical Information Service, Springfield, VA.
- Johnson, W. E. and J. R. Vallentyne. 1971. Rationale, background and development of experimental lake studies in Northwestern Ontario. J. Fish. Res. Bd. Canada 28 (2): 123-128.

- Kaesler, R. L. and J. Cairns, Jr. 1972. Cluster analysis of data from limnological surveys of the upper Potomac River. Amer. Mid. Nat. 88 (1): 56-57.
- Lighthart, B. and H. Bond. 1976. Design and preliminary results from soil/litter microcosms. Intern. J. Environmental Studies 10: 51-58.
- National Academy of Sciences. 1975. Principles for Evaluating Chemicals in the Environment (Washington D.C.: National Academy of Sciences).
- National Research Council. 1979. Commission on Natural Resources. Committee to Review Methods in Ecotoxicology. Minutes. 25-26 October.
- Taub, F. B. 1976. Demonstration of pollution effects in aquatic microcosms. Intern. J. Environmental Studies 10: 23-33.
- Whittaker, R. H. 1961. Experiments with radiophosphorous tracer in aquarium microcosms. Ecol. Monogr. 31 (2): 157-186.



An Ecosystem Approach to the Toxicology of  
Residue Forming Xenobiotic Organic  
Substances in the Great Lakes

A Manuscript Invited by  
The Environmental Studies Board  
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The ubiquitous distribution of residue forming xenobiotic substances, particularly organochlorine compounds, is rapidly becoming apparent. Widespread dissemination is suggested by a variety of studies of areas presumably remote from the direct industrial and/or cultural influences attributable to man, including melting snow in the Antarctic (Peterle, 1969; Peel, 1975), mammals of the arctic (Bowes and Jonkel, 1975; Clausen *et al.*, 1975); in the surface waters and atmosphere above the Sargasso Sea (Bidleman and Olney, 1974); the waters of the Gulf of Mexico (Giam *et al.*, 1973); remote island sites (Swain, 1978); rainfall over the Hawaiian Islands (Benvenue *et al.*, 1972) and the sediments of Siberian Lake Baikal (Swain, 1980). Although a number of these compounds have been banned outright, or had their utilization severely restricted in the 1969 to 1971 time period, they still are routinely reported in the Great Lakes Basin (International Joint Commission, 1977; 1978). Strachan and Huneault (1979) note three principal reasons for this continued occurrence: 1) the widespread geographic occurrence of organochlorine substances; 2) their persistence and relatively long biological half-lives; and 3) the demonstration of long range atmospheric transport of these substances.

The persistence of many of the xenobiotic compounds of concern is measured in years or decades (International Joint Commission, 1975). This fact, combined with hydraulic detention times of the order of centuries for the sequentially coupled St. Lawrence Great Lakes (Rainey, 1967), yields a potential for bioaccumulation and bioconcentration virtually unparalleled elsewhere in North America. When one considers that the Great Lakes represent 20 percent of the world's freshwater and 95 percent of the freshwater by volume of the adjacent United States (McGrath, 1980), the problem is of even greater interest.

Because of the complexity of the issue; the multiplicity of substances and their metabolites, daughter compounds, and degradation products; the size and scope of the ecosystem in both spatial and temporal scales; and the intricacy and interdependence of the trophic system in the Great Lakes, it was reasoned that an ecosystem approach to the problem was the method of choice for examining the relationships and proportions of these interdependent entities. The use of such a conceptual framework related to toxic substances has been previously suggested (Gillett *et al.*, 1974; Metcalf *et al.*, 1975; Thomann, 1978, 1980; Bierman and Swain, 1978; and Weininger, 1978). However, few of these approaches have considered man as an integral part of the environmental structure, and none have regarded him as both an interactive and reciprocal part of the ecosystem.

The intent of the present study is to consider the broader sphere of influence of toxic organic substances upon the ecosystem from input sources through top level consumers, including man. To do so requires the use of an individual organochlorine compound or class of such substances capable of serving as indicators or tracer substances which fit the following criteria:

1. The compound must be ubiquitous in nature and of wide geographic distribution,
2. The size of the reservoir of this substance outside of the aquatic ecosystem should be large, with demonstrated transport mechanisms to the water mass,
3. The compound should be poorly metabolized and biodegraded so that it will behave as conservatively as possible,
4. It should have a demonstrated lipophilicity, and hence will bioaccumulate and bioconcentrate within the ecosystem, and
5. Ideally, as a class of compounds serve as a potential indicator or surrogate for predicting the behavior of other materials based on varying physiochemical structure/biological activity correlations.

In the Great Lakes ecosystem, the substances which most closely fulfill these requirements are the polychlorinated biphenyl compounds (PCBs). Thus, while other specific compounds will be considered as contributing to the organochlorine burden of the Great Lakes, PCBs will be utilized as an surrogate for many of these other substances in the proposed ecosystem conceptual framework. It is intended that the use of these substances will provide insight into the ecosystem behavior of toxic substances in the Great Lakes.

Figure 1 represents an attempt to demonstrate the major compartments which contribute substantially to the movement of toxic substances through the ecosystem of the Great Lakes, particularly the upper lakes, Superior, Michigan and Huron. The upper layers of this figure suggest the major pathways and mechanisms which are important in both the distribution of persistent residue forming xenobiotic compounds and in their potential impact upon human populations. While an exhaustive treatment of each ecosystem compartment is beyond the scope of this effort, the intent is to provide a single holistic concept into which precise quantifiable data may be inserted for a wide variety of toxic substances and disseparate ecosystems. Thus, a variety of work is summarized for each compartment, and, where available, reference is made to current definitive works pertinent to the problem. The remainder of this study, therefore, will consider the interrelationships and proportions among the various entities represented in the conceptual framework related to toxic substances.

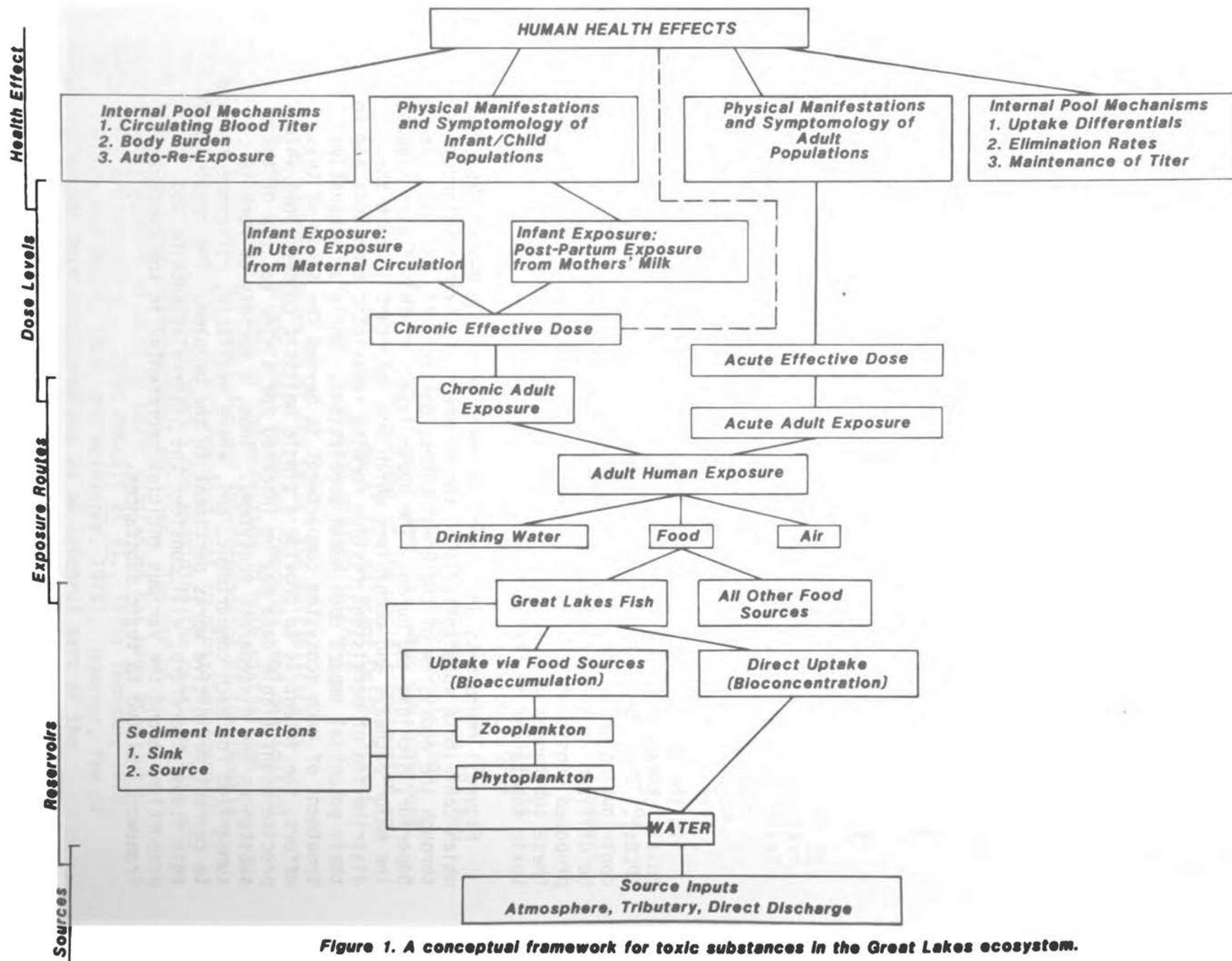


Figure 1. A conceptual framework for toxic substances in the Great Lakes ecosystem.

### Source Inputs of Xenobiotic Compounds

The sources for addition of organochlorine substances to the Great Lakes are similar to those of other polluting substances, i.e., atmospheric inputs, tributary loading, direct discharges, and other diffuse sources. However, in the case of many of the organochlorine compounds, the relative importance of each of these source terms is significantly different from reported data on other polluting substances, e.g., trace metals and nutrient parameters effecting eutrophication. For those lakes upon which substantial effort has been expended to obtain annual net fluxes of organochlorine compounds, e.g., Lakes Superior and Michigan, present best estimates suggest the bulk of the contribution of these substances to the lakes are from atmospheric sources. Eisenreich (1980a) has indicated that as much as 90 to 95 percent of the PCB loading to Lake Superior may be the result of atmospheric contributions. Murphy (1980) has reported similar findings for Lake Michigan; an atmospheric contribution of greater than 75 percent. These workers suggest mean values for ambient atmospheric contamination by PCB at a level of approximately  $1.5 \text{ ng/m}^3$  to air over Lake Superior (Eisenreich, 1980) and between 5 and  $10 \text{ ng/m}^3$  over Lake Michigan (Murphy, 1980). Rice (1980) has found ambient atmospheric concentrations of PCBs over Lake Michigan ranging from 0.96 to  $5.37 \text{ ng/m}^3$ , with a definite increase in concentrations along the north-south axis of the lake as the metropolitan regions of the south are approached. These findings are confirmed by Murphy (1980), who notes an increase in excess of 100 fold in PCB loading values to Lake Michigan adjacent to Chicago, as compared with the northern portions of the lake near the Straits of Mackinac. Andren and Doskey (1977) have found  $0.12 \text{ ng/m}^3$  PCBs in filtered extracts of air over Lake Michigan, of which 76 percent was Aroclor 1242 and 24 percent was Aroclor 1254. They also observed  $1.4 \text{ ng/m}^3$  of PCBs in the vapor state in a ratio of 70 percent Aroclor 1242 to 30 percent Aroclor 1254.

Little is known about ambient concentrations in the atmosphere of other organochlorine compounds. In fact, a review of the literature indicates a single study of the Great Lakes region providing data on this question. Stanley, *et al.* (1971) reported values for atmospheric pesticides. Their work shows p,p'-DDT at  $11.0 \text{ ng/m}^3$ , and  $2.9 \text{ ng/m}^3$  of o,p'-DDT in air samples over Lake Erie near Buffalo, New York.

A substantial data set for the occurrence of PCB in atmospheric precipitation exists, however. These data are summarized in Table 1. As in the case of ambient atmospheric studies, the available knowledge on other organochlorine compounds in Great Lakes precipitation is limited. The International Joint Commission (1978) reporting rainfall data from their Pollution from Land Use Activities Reference Group (PLURG) observed ranges of inputs for p,p'-DDT to Lake Erie between 4 and  $16 \text{ ng/l}$ ; for Lake St. Clair between not detectable and  $7 \text{ ng/l}$ ; for Lake Huron between not detectable and  $13 \text{ ng/l}$ ; and for Lake Ontario between 3 and  $19 \text{ ng/l}$ . Heptachlor epoxide was also found by these workers in rainfall taken adjacent to Lake St. Clair, but levels were below the quantification

TABLE 1

PCB's in Precipitation in the Great Lakes Basin

<u>LAKE</u>	<u>Site</u>	<u>Mode</u>	<u>Quantity ng/g</u>	<u>Reference</u>
<u>LAKE ERIE</u>	Essex County Ontario	Rainfall	30-70	Ontario Ministry of the Environment (1976)
	Open Lake	Rainfall	9	Strachan (1978)
	Lake Erie Watershed	Rainfall	10-100	Frank et al. (1978)
	Lake Erie Watershed	Rainfall	10-100	Sanderson (1977)
	Lake Erie Watershed	Rainfall	9	Strachan & Huneault (1979)
<u>LAKE ST. CLAIR</u>	Lake St. Clair Watershed	Rainfall	20-100	Frank et al. (1978)
	Lake St. Clair Watershed	Rainfall	10-70	Sanderson (1977)
<u>LAKE HURON</u>	Lake Huron Watershed	Rainfall	10-100	Frank et al. (1978)
	Lake Huron Watershed	Rainfall	40-100	Sanderson (1977)
	Saginaw Bay	Rainfall	19	Murphy (1978)
	Georgian Bay	Rainfall	11	Strachan (1978), Strachan & Huneault (1979)
	Georgian Bay	Snowfall	18	Strachan & Huneault (1979)
<u>LAKE SUPERIOR</u>	Open Lake	Rainfall	26	Strachan (1978)
	Western Lake Superior	Snowfall	50	Swain (1978)
	Isle Royale	Snowfall	230	Swain (1978)
	Lake Superior Watershed	Snowfall	38	Strachan & Huneault (1979)
	Lake Superior Watershed	Rainfall	40	Strachan & Glass (1978)
<u>LAKE MICHIGAN</u>	Chicago	Rainfall	193	Murphy (1978), Murphy & Rzeszutko (1977)
	Chicago	Snowfall	212	Murphy (1978), Murphy & Rzeszutko (1977)
	Beaver Island	Rainfall	215	Murphy (1978), Murphy & Rzeszutko (1977)
<u>LAKE ONTARIO</u>	Lake Ontario Watershed	Rainfall	nd-100	Ontario Ministry of the Environment (1976)
	Lake Ontario Watershed	Rainfall	20	International Joint Commission (1977)
	Lake Ontario Watershed	Rainfall	32	Strachan & Huneault (1979)
	Lake Ontario Watershed	Snowfall	43	Strachan & Huneault (1979)

threshold. Unquestionably, however, the most complete data set for organochlorine compounds other than PCB in precipitation in the Great Lakes basin is provided by Strachan and Huneault (1979). Their results are summarized in Table 2.

This detailed effort followed a preliminary identification in 1976 of a variety of organochlorine substances as routine contaminants in rain water in the Lake Ontario area (International Joint Commission, 1977). In addition to PCBs, these substances included:

Lindane	DDT residues
Methoxychlor	$\alpha, \beta$ Endosulfan
Dieldrin, and cis-, trans-chlordane	

In this study, Mirex and BHC were also tentatively identified.

As the state of knowledge of atmospheric transport and removal processes advances, calculations of relative burden and contributions to aquatic ecosystems can be made. Using a similar approach to the calculation of net deposition of atmospheric contaminants outlined for the South Carolina coast by Bidleman and Christensen (1979a, 1979b), Doskey (1978) calculated atmospheric loading values for PCBs in Lake Michigan. He reported observed mean concentrations of PCBs at 1.0 ng/m<sup>3</sup>. Particulate phase PCB values made up approximately 13 percent of the total particulate concentrations and were observed in the 0.09 to 0.37 ng/m<sup>3</sup> range. Using both a liquid and a gas phase control models, the wet and dry flux of PCBs to Lake Michigan were estimated to be 1101 kg/yr and 3354 kg/yr, respectively. The dry flux of vapor phase PCBs was calculated at 2165 kg/yr, while a re-volatilization rate of 3132 kg/yr PCB was estimated. Using the measured wet flux value of Murphy and Rzeszutko (1977) of 4800 kg/yr, net atmospheric fluxes under the gas and liquid phase models were calculated at 8655 kg/yr and 2848 kg/yr, respectively.

Hollod (1979) has carried the application of mathematical modeling of toxic xenobiotic compounds in the Great Lakes one step further. His work provides an input-output model for Lake Superior for polychlorinated biphenyl compounds. His mass balance projections suggest a surplus in inputs over loss terms of 6400-8700 kg/yr of PCB (see Table 3). Aside from graphically demonstrating the relative importance of each of the contributing factors, e.g., atmosphere, diffuse sources, municipal effluents, industrial discharge, etc., the calculation clearly shows a large net accumulation of toxic substances in the water column, suggesting that the lake has yet to reach equilibrium with its burden. Eisenreich (1980a) estimates that the calculated excess of sources over loss terms will result in a net accumulation in the water column which will increase ambient concentrations in Lake Superior by approximately 0.2 ng/l/yr.

At the very least, this study suggests that the total impact from compounds which have been banned for a considerable period is yet to be felt. For a further discussion of the question of the importance of atmospheric sources, the reader is referred to Eisenreich *et al.* (1980b).

TABLE 2

Mean Concentrations of Toxic Substances and Pesticides in Rain and Snowfall.<sup>1</sup> Values Expressed in ng/l; nd = not detected.

	<u>Rainfall</u>				<u>Snowfall</u>				
	Lake Superior	Lake Huron Georgian Bay	Lake Ontario	Lake Erie	Lake Superior	Lake Huron Georgian Bay and North Channel	Eastern and Central Ontario	North Eastern Ontario	All Snow Samples
Lindane	4.9	6.0	4.7	6.1	0.1	nd	0.4	nd	0.1
α - BHC	4.6	13.3	19.1	10.3	1.5	0.8	0.5	0.8	0.9
Σ DDT Residues	0.8	2.7	5.6	3.8	1.9	0.4	1.5	0.3	1.0
α - Endosulfan	0.2	0.1	3.8	1.6	nd	nd	nd	nd	nd
β - Endosulfan	1.0	2.1	12.0	2.0	nd	nd	0.1	nd	nd
Dieldrin	0.5	1.0	1.3	2.6	nd	nd	nd	nd	nd
Methoxychlor	1.6	9.5	8.5	13.1	0.1	0.2	5.8	0.3	1.5
HCB	2.8	nd	nd	nd	nd	0.1	nd	nd	nd

<sup>1</sup>From Strachan and Huneault, 1979.

TABLE 3

Mass Balance of PCB in Lake Superior<sup>1</sup>

<u>Input Terms</u>		<u>Output Terms</u>	
<u>Source</u>	<u>Value in kg/yr.</u>	<u>Source</u>	<u>Value in kg/yr.</u>
1. Atmosphere	6600-8300	1. St. Mary's River	140
2. Tributaries	<1311	2. Sedimentation	1000-1600
3. Municipal	66		
4. Industrial	2		
	<hr/>		<hr/>
Total	~ <8000-9700	Total	1140-1740

Net accumulation = excess of inputs over losses: 6400-8700 kg/yr.

<sup>1</sup>From Hollod, G.J. (1979)

### Accumulations in Great Lakes Water Masses, Plankton, and Sediment

A review of extant data relative to organochlorine compounds in the waters of the Great Lakes shows a high variability in comparisons made both between lakes and within a single given body of water. This variation for polychlorinated biphenyl compounds is described in Table 4. The lowest reported value for Great Lakes waters was that of Veith *et al.* (1977), who reported a value of 0.0008  $\mu\text{g}/\ell$  (0.8  $\text{ng}/\ell$ ) PCB as Aroclor T254 in a 1972-73 sample of water taken from the lake water intake of the Environmental Protection Agency's Environmental Research Laboratory-Duluth, at Duluth, Minnesota. Maximum reported levels of PCB in the Great Lakes are to be found in nearshore samples of Lake Huron waters adjacent to tributary streams. Values reported for these water masses by the Michigan Department of Natural Resources to the International Joint Commission fell between 0.4 and 0.7  $\mu\text{g}/\ell$  (400-700  $\text{ng}/\ell$ ) PCB (International Joint Commission, 1978). The wide range of values for the water masses of the Great Lakes is apparently indicative of a general lack of definitive work in this area, largely as a function of two principal factors:

1. The high degree of variability of organochlorine compounds in the waters of the Great Lakes, both temporally and spatially, and
2. Only recently has the analytical capability and methodology progressed to the point where nanogram per liter concentrations can be adequately and reliably separated from potential interference by these ubiquitous compounds.

It is currently held that ambient average concentrations for the open water masses of the Great Lakes are in a general range of 1 to 10  $\text{ng}/\ell$ , while nearshore values and areas adjacent to tributary inputs may be substantially higher. Further, Eisenreich (1980a) has demonstrated in Lake Superior that a sharply diminishing gradient exists within the water column from the surface down to 25 meters, suggesting an enrichment of the surface from atmospheric deposition. He reports ambient average concentrations for lake water concentrations of PCB in the open waters to be approximately 1.0  $\text{ng}/\ell$ . He notes surface concentrations in a range of 0.4 to 7.4  $\text{ng}/\ell$  (mean, 1.9  $\text{ng}/\ell$ ) in Lake Superior, while epilimnetic concentrations near the bottom are rather consistently of the order of 1.0  $\text{ng}/\ell$ . Both Mount (1976) and Swain (1978) report values for the open Lake Superior at 5.0  $\text{ng}/\ell$ , the former in deep waters adjacent to a barrel dump site, the latter in the surface waters.

The influence of nearshore effects is suggested by Strachan and Glass (1978), who, reporting STORET data, note 50 to 150  $\text{ng}/\ell$  PCB in the shoreward waters of Lake Superior. Swain (1978) reports that in the confined embayments of Isle Royale under ice cover, surface concentrations of PCB reached 0.120 to 0.157  $\mu\text{g}/\ell$  (120-157  $\text{ng}/\ell$  PCB).

Polychlorinated biphenyl values for the other Great Lakes are apparently fewer, and more highly variable (Table 4). In Saginaw Bay (Lake Huron), Kinkead and Hamdy (1978) observed a range of 0-10 ng/l PCB for the Outer Bay, 13-23 ng/l for the Inner Bay, and 80-90 ng/l PCB adjacent to the Saginaw River.

Glooschenko *et al.* (1976) report 27.0 ng/l as a lakewide mean value for Lake Erie 1972 data. The only other comparable study available reviewed 1974-75 data, noting values below a 20.0 ng/l quantification limit (Pennsylvania Department of Natural Resources, 1977). Data for various nearshore stations on Lake Ontario are reported by the International Joint Commission (1977) as falling in a range of 44.0 to 77.0 ng/l PCB.

Similarly, published data for Lake Michigan includes only values for nearshore stations. Torrey (1976) reports a range of nearshore values of 12-56 ng/l while Murphy and Rzeszutko (1977) observed a range of 30-40 ng/l in the waters adjacent to Beaver Island and Chicago, respectively. Recent work by Armstrong (1980) and by Rice (1980) suggest that the lakewide mean for the open surface waters of Lake Michigan will fall between 5 and 10 ng/l, probably approaching 7 ng/l PCB.

A wide variety of other xenobiotic organochlorine compounds have been observed in the Great Lakes. The International Joint Commission (1977, 1978) has reported identification of more than 300 of these compounds. However, certain of these compounds are repeatedly observed in most, if not all, of the waters of the Great Lakes. Included in this group are the following:

Aldrin	Heptachlor
Dieldrin	Heptachlor Epoxide
Endrin	Chlordane
Lindane	Methoxychlor
DDT and its Metabolites	Phthalic Acid Esters

While an exhaustive description of the distribution of these substances is beyond the scope of this effort, a summary of observations of these substances is provided in Table 5.

If water column values of organochlorine compounds are few, data for concentrations in benthic and planktonic constituents are virtually non-existent. The most extensive data sets for the Great Lakes literature are available for Lake Superior and Lake Ontario. Glooschenko *et al.* (1976), working on Superior, observed PCBs in 7 of 15 seston samples in concentrations ranging from 0.5 to 1.3 mg/kg. Of the eight remaining samples, three had levels that were below the threshold of detection, while the remaining five were detectable, but below the level of quantification. Trace quantities of dieldrin were also found in most of the benthic samples taken. A single data set for the planktonic constituents has been reported for Lake Superior. Veith *et al.* (1977) report from 0.05 to 0.12 mg/kg (ppm) PCB, and 0.04 to 0.05 mg/kg (ppm) of DDT residues and 2.0 µg/kg (ppb) of dieldrin in the samples of the opossum shrimp *Mysis relicta* taken from the Minnesota north shore of western Lake Superior during 1973-1974.

TABLE 4  
 POLYCHLORINATED BIPHENYL COMPOUNDS OBSERVED  
 IN WATERS OF THE GREAT LAKES

Location	Date	Quantity in ng/l	Reference
<u>Lake Superior</u>			
Open Waters Western Arm	1976	5.0	Mount, 1976
Open Lake	1974	5.0	Swain, 1978
Nearshore Western Arm	1974	7.0	Swain, 1978
Nearshore Western Arm	1972	0.8	Veith <u>et al.</u> , 1977
Nearshore Western Arm Under Ice Cover	1974	10.0	Swain, 1978
Duluth Superior Harbor Howard's Bay-Under Ice Cover	1974	20.0	Swain <u>et al.</u> , 1975
Duluth Superior Harbor Howard's Bay-Under Ice Cover	1974	29.0	Swain <u>et al.</u> , 1975
Michigan Nearshore	1971-75	150	STORET in Strachan & Glass, 1978
Wisconsin Nearshore	1974-75	50	STORET in Strachan & Glass, 1978
Minnesota Nearshore	1972-75	110	STORET in Strachan & Glass, 1978
<u>Lake Michigan</u>			
Various Nearshore Waters	1976	12-56	Torrey, 1976
Nearshore, Vicinity of Chicago	1977	40	Murphy and Rzeszutko, 1977
Nearshore, Beaver Island	1977	30	Murphy and Rzeszutko, 1977

TABLE 4 (Cont.)

Location	Date	Quantity in ng/l	Reference
<u>Lake Huron</u>			
Saginaw Bay Saginaw River	1974	80-90	Kinkead and Hamdy, 1978
Saginaw Bay Inner Bay	1974	13-23	Kinkead and Hamdy, 1978
Saginaw Bay Outer Bay	1974	0-10	Kinkead and Hamdy, 1978
Nearshore, Vicinity of Cheboygan River	1973-76	500.0	Mich. Dept. of Nat. Res. in IJC, 1978
Nearshore, Vicinity of Rifle River	1972-75	700.0	Mich. Dept. of Nat. Res. in IJC, 1978
Nearshore, Vicinity of Oscoda	1973-76	400.0	Mich. Dept. of Nat. Res. in IJC, 1978
<u>Lake Erie</u>			
Lakewide Mean	1972	27.0	Glooschenko <i>et al.</i> , 1976
Nearshore, Vicinity of Erie, PA	1974-75	20.0	Pennsylvania Dept. of Nat. Res. in IJC, 1978
<u>Lake Ontario</u>			
Various Nearshore Stations	1972-73	44.0 to 77.0	IJC, 1977
<u>St. Lawrence River</u>			
15 samples in St. Lawrence River, locations undefined	1971	300	Dennis, 1975
29 samples in St. Lawrence River, locations undefined	1972	100	Dennis, 1975
31 samples in St. Lawrence River, locations undefined	1973	100	Dennis, 1975
38 samples in St. Lawrence River, locations undefined	1974	ND	Dennis, 1975

TABLE 5  
 ORGANOCHLORINE SUBSTANCES OBSERVED IN THE WATERS  
 OF THE GREAT LAKES

Location	Contaminant	Quantity in $\mu\text{g}/\ell$	Date	Reference
<u>Lake Superior</u>				
Michigan Nearshore	Diethylhexyl Phthalate	2.0	1976	Mich. Dept. of Nat. Res., 1978
Michigan Nearshore	o,p-DDT	0.001-0.07	1973-1974	Mich. Dept. of Nat. Res., 1978
Michigan Nearshore	p,p-DDT	0.02-0.025	1974	Mich. Dept. of Nat. Res., 1978
Open Waters	Lindane	trace	1974	Glooschenko, <u>et al.</u> , 1976
Duluth-Superior Harbor	Lindane	0.002	1974	Swain, <u>et al.</u> , 1975
Duluth-Superior Harbor	BHC	0.005	1974	Swain, <u>et al.</u> , 1975
Michigan Nearshore	Aldrin + Dieldrin	0.040	1973-1975	Strachan & Glass, 1978
Wisconsin Nearshore	Aldrin + Dieldrin	0.008	1974-1975	Strachan & Glass, 1978
Michigan Nearshore	Total DDT Residues	0.063	1971-1975	Strachan & Glass, 1978
Wisconsin Nearshore	Total DDT Residues	0.022	1974-1975	Strachan & Glass, 1978
Minnesota Nearshore	Total DDT Residues	0.032	1967-1975	Strachan & Glass, 1978
Michigan Nearshore	Heptachlor & Heptachlor Epoxide	0.038	1973-1975	Strachan & Glass, 1978
Wisconsin Nearshore	Heptachlor & Heptachlor Epoxide	0.005	1974-1975	Strachan & Glass, 1978
Michigan Nearshore	Lindane	0.008	1973-1975	Strachan & Glass, 1978

TABLE 5 (Cont.)

Location	Contaminant	Quantity in $\mu\text{g}/\ell$	Date	Reference
<u>Lake Superior, Cont.</u>				
Wisconsin Nearshore	Lindane	0.002	1974- 1975	Strachan & Glass, 1978
Michigan Nearshore	Endrin	0.043	1973- 1975	Strachan & Glass, 1978
Michigan Nearshore	Chlordane	0.059	1973- 1975	Strachan & Glass, 1978
Wisconsin Nearshore	Methoxychlor	0.015	1974- 1975	Strachan & Glass, 1978
<u>Lake Michigan</u>				
Open Lake Waters	Lindane	0.0006-0.0019	1969- 1970	Lake Michigan Enforcement Conference, 1972
Open Lake Waters	Heptachlor	0.0007-0.0031	1969- 1970	Lake Michigan Enforcement Conference, 1972
Open Lake Waters	Aldrin	0.0006-0.0019	1969- 1970	Lake Michigan Enforcement Conference, 1972
Open Lake Waters	Endrin	0.0021-0.0053	1969- 1970	Lake Michigan Enforcement Conference, 1972
Open Lake Waters	Dieldrin	0.001-0.0042	1969- 1970	Lake Michigan Enforcement Conference, 1972
Open Lake Waters	Total DDT	0.003 to 0.0282	1969- 1970	Lake Michigan Enforcement Conference, 1972

TABLE 5 (Cont.)

Location	Contaminant	Quantity in $\mu\text{g}/\ell$	Date	Reference
<u>Lake Michigan, Cont.</u>				
Michigan Nearshore	DDT	0.002	1968	Torrey, 1976
Michigan Nearshore	DDT	0.001	1968	Torrey, 1976
Michigan Nearshore	DDE	0.0005	1968	Torrey, 1976
Michigan Nearshore	Dieldrin	0.001	1968	Torrey, 1976
Michigan Nearshore	Dibutyl Phthalate	1.3	1975- 1976	Mich. Dept. of Nat. Res., 1978
Michigan Nearshore	Diethylhexyl Phthalate	1.5-3.0	1975- 1976	Mich. Dept. of Nat. Res., 1978
Illinois Nearshore	Total DDT	0.0003-0.0004	1970	Schacht, 1974
<u>Lake Huron</u>				
Michigan Nearshore	DDT	0.001-0.004	1974	IJC, 1977
Open Lake	Lindane	Trace	1974	Glooschenko, <u>et al.</u> , 1976
Open Lake	Heptachlor	Trace	1974	Glooschenko, <u>et al.</u> , 1976
Open Lake	Dieldrin	Trace	1974	Glooschenko, <u>et al.</u> , 1976
Saginaw Bay	Dibutyl Phthalate	1.0	1974	Ewing, 1977
Saginaw Bay	Diethylhexyl Phthalate	1.0	1974	Ewing, 1977
Saginaw Bay	Dieldrin	500-600	1974	Kinthead and Hamdy, 1978
Michigan Nearshore	Diethylhexyl Phthalate	0.73-18.0	1973- 1977	Mich. Dept. of Nat. Res., 1978

TABLE 5 (Cont.)

Location	Contaminant	Quantity in $\mu\text{g}/\ell$	Date	Reference
<u>Lake Huron, Cont.</u>				
Michigan Nearshore	Dibutyl Phthalate	0.6-8.6	1973-1977	Mich. Dept. of Nat. Res., 1978
Michigan Nearshore	p,p-DDT	0.11-0.024	1973-1977	Mich. Dept. of Nat. Res., 1978
Michigan Nearshore	o,p-DDT	0.01	1973-1977	Mich. Dept. of Nat. Res., 1978
<u>Lake Erie</u>				
Open Waters	Phthalate Esters	0.7-6.0	1973	Strachan, 1976
Michigan Nearshore	Diethylhexyl Phthalate	2.6	1975-1976	Mich. Dept. of Nat. Res., 1978
Selected Nearshore Areas	Pentachlorophenol	0.005-1.7	1977	Fox, 1978
Selected Nearshore Areas	Total DDT	ND-0.347	1975-1977	IJC, 1978
Selected Nearshore Watersheds	Dieldrin	ND-0.120	1975-1977	IJC, 1978
Selected Nearshore Watersheds	Chlordane	ND-0.004	1975-1977	IJC, 1978
Selected Nearshore Watersheds	Heptachlor Epoxide	ND-0.37	1975-1977	IJC, 1978
<u>Lake Ontario</u>				
Open Lake Waters	Lindane	<0.005-0.008	1976	IJC, 1977
Open Lake Waters	Dieldrin	0.0013-0.012	1975	IJC, 1977
Open Lake Waters	DDE	0.0094-0.045	1975	IJC, 1977
Open Lake Waters	DDD	<0.0005-0.013	1975	IJC, 1977
Open Lake Waters	DDT	0.0014-0.012	1975	IJC, 1977
Open Lake Waters	Total DDT	0.016-0.057	1975	IJC, 1977

A single report for organochlorine compounds in the plankton and benthos is presently available for Lake Michigan. Veith (1973) reported PCB levels in the amphipod *Pontoporeia affinis* for two sites in the Lake Michigan nearshore environment. He observed 0.06 mg/kg in this occasionally planktonic form near Sturgeon Bay, and 0.45 mg/kg near Waukegon.

In Lake Huron, Glooschenko *et al.* (1976) reported PCB in all but one sample of seston taken. The range of values found by these workers was 0.5 to 8.1 mg/kg. Trace quantities of dieldrin and p,p'-DDE were also routinely found.

For Lake Erie, useful values are equally limited. The Great Lakes Research Institute (1973) reported trace amounts of an unidentified Arochlor in the benthos of Presque Isle Bay, and in a 1974 report, this group reported observing DDT, lindane, aldrin, dieldrin and heptachlor in benthic forms from this same area.

The International Joint Commission (1977), reporting International Field Year on the Great Lakes (IFYGL) data for Lake Ontario, noted the occurrence of DDT and its metabolites, dieldrin and PCB in plankton, *Cladophora* sp., an attached benthic algal form, and in benthic fauna. These values are summarized in Table 6.

More recent studies (1975) have also been reported by the International Joint Commission (1977). In these efforts by staff of the Canada Centre for Inland Waters, 64  $\mu$  mesh net plankton were collected at 11 stations. Residues of DDT and its metabolites, dieldrin and PCBs were found at all stations (Table 7). Again in this instance, highest observed values were reported from the nearshore areas, principally the Niagara plume, adjacent to Oswego, New York, and in Hamilton Harbor. It should be noted, however, that it cannot be determined for the values reported whether a wet weight or dry weight basis was used, since a notation of both labels appears on the data.

The studies undertaken by McNaught and his co-workers (1978) suggest that efforts toward a better understanding of the underlying mechanisms associated with organochlorine compounds and their impact on Great Lakes planktonic constituents may be critical to our knowledge of ecosystem impacts of these compounds. These researchers report a substantial reduction in primary productivity upon exposure to PCB mixtures, and recent additional evidence (McNaught, 1980) demonstrates an enhancement of the dark adaptation phenomenon. This latter observation has also been confirmed by Rhee (1980). It is clear that additional efforts are needed in this important portion of trophic hierarchy.

With respect to polluting substances, the sediments of the Great Lakes have been shown to act both as a sink (Kinkead and Chatterjee, 1974; Eisenreich *et al.*, 1979; Smith *et al.*, 1980; Eisenreich and Johnson, 1980; Glooschenko *et al.*, 1976; Maile, 1977; Veith *et al.*, 1977; Torrey, 1976; Frank *et al.*, 1979; Kleinert, 1976; and Dennis, 1976) and as a source of contaminants (Lick, 1979; Eisenreich, 1980; Hollod, 1979; Dennis, 1976; DiToro, 1974; Sydor *et al.*, 1978; Sonzogni *et al.*, 1980;

TABLE 6

OCCURRENCE OF CHLORINATED HYDROCARBONS IN THE PLANKTON,  
CLADOPHORA, AND BENTHIC FAUNA OF LAKE ONTARIO<sup>1</sup>

	<u>Net Plankton</u> μg/g <u>Dry Weight</u>	<u>Cladophora</u> ng/g <u>Dry Weight</u>	<u>Benthos</u> ng/g <u>Dry Weight</u>
DDE	1.19-5.89	97-347	26-124
DDD	<0.05-0.37	0.45-30	1.8-26
DDT	<0.05-0.86	1.1-16	2.4-59
TOTAL DDT RESIDUES	1.2-5.9	119-365	32-209
DIELDRIN	0.05-0.25	1.9-25	3.0-14.8
PCB	N.D.-11.8	333-860	97-976

<sup>1</sup>From International Joint Commission, 1977

TABLE 7

CONCENTRATIONS ( $\mu\text{g/g}$  dry weight) OF CHLORINATED HYDROCARBONS  
IN LAKE ONTARIO NET PLANKTON ( $64 \mu$  mesh) - 1975<sup>1</sup>

<u>Contaminant</u>	<u>No. of Samples</u>	<u>% Sample Exceeding Detection Limit</u>	<u>Minimum Value</u>	<u>Maximum Value</u> ( $\mu\text{g/g}$ wet weight)	<u>Mean Value</u>
Aldrin/Dieldrin	11	91	0.010	0.41	0.136
Chlordane	11	18	0.031	0.72	0.37
DDT and Metabolites	11	91	0.094	1.26	0.376
Endrin	11	0	less than 0.01, detection limit		
Heptachlor	11	0	less than 0.001, detection limit		
Heptachlor Epoxide	11	27	0.008	0.094	0.038
Lindane	11	27	0.006	0.021	0.012
Methoxychlor	11	0	less than 0.05, detection limit		
PCBs	11	91	0.4	6.3	1.88

<sup>1</sup>From International Joint Commission (1977)

Kang and Lick, 1980; Sly and Sandilands, 1980; and Richardson et al., 1980). Not only has resuspension been demonstrated to be of importance by these authors, Weininger (1978) has shown that organochlorine compounds may be recycled by the biologic community of the sediments, thus inhibiting permanent burial.

A strong association of many of the xenobiotic organochlorine compounds with suspended solids has been demonstrated (Eisenreich et al., 1979; Hallod, 1979; Richardson, 1980; Mullin and Filkins, 1980; Ulanoff et al., 1980; Erstfeld et al., 1980). The association with suspended particulates is of such a character, that it enables the use of the net settling velocity of these materials as an internal loss mechanism for particulate sorbed organochlorine compounds in ecosystem mass conservation models (Thomann, 1978; Bierman and Swain, 1978; Richardson, 1980). Hollod (1979) has summarized the mass sedimentation rates for the five Great Lakes. His work is shown in Table 8. This author and Eisenreich et al. (1979) have calculated net accumulation rates for Lake Superior. They have shown deposition rates to vary over two orders of magnitude (0.01 - 0.11 cm/yr). Eisenreich (1980) has observed a mean concentration of 0.13  $\mu\text{g/g}$  (dry weight basis) of PCB associated with the surficial sediments of Lake Superior, from which a loss to the sediment of 1000 - 1600 kg/yr PCB is calculated (Eisenreich, 1980; Hollod, 1979). These workers further note that sedimentary concentrations of PCBs increase with increasing sedimentation rates.

A variety of studies have examined the concentrations of organochlorine substances in the sediments of the Great Lakes. A synopsis of these efforts is contained in Table 9. Among the most definitive efforts of sedimentary accumulation in the Great Lakes are the studies of Thomas and his co-workers. For a more detailed description of sedimentary accumulation, the reader is referred to Thomas et al. (1979).

#### Accumulation in Great Lakes Fish

Because of their widespread availability throughout the Great Lakes ecosystem, their persistence, their lipophilicity, and the long hydraulic detention times of the coupled system, toxic organochlorine compounds of anthropogenic origin tend to bio-magnify in Great Lakes biota; either through food chain transfer (bioaccumulation) or as a function of direct uptake across gill or other surface membranes (bioconcentration). While there is some question as to the exact magnitude of contribution of each of these phenomena to the body burden of Great Lakes fish (Weininger, 1978, estimates approximately 95 percent food chain transfer and 5 percent direct uptake; Thomann, 1978, calculates 75 percent bioaccumulation and 25 percent bioconcentration) there is no argument that the higher trophic levels show substantially increased levels of organochlorine contaminants as compared with the water masses, the primary producers and the primary consumers.

TABLE 8  
 MASS SEDIMENTATION RATES IN THE GREAT LAKES<sup>1</sup>

Lake	Mass Sedimentation Rate mg/cm <sup>2</sup> /yr    cm/yr		Reference
Superior	2.5-78	0.1-0.2	Kemp <u>et al.</u> , 1978 Robbins <u>et al.</u> , 1978
	0-88	0-0.11	Eisenreich <u>et al.</u> , 1979
Michigan	6-102	0.01-0.4	Robbins & Edgington, 1975 Robbins, 1978
Huron	3.5-98	0.02-0.33	Kemp and Harper, 1977
Erie	13-204	0.02-0.85	Nriagu <u>et al.</u> , 1979 Kemp <u>et al.</u> , 1978
Ontario	8.5-122	0.03-0.12	Kemp <u>et al.</u> , 1978

<sup>1</sup>Data from Hollod, G.J. (1979)

TABLE 9. ORGANOCHLORINE COMPOUNDS IN THE SEDIMENTS OF THE GREAT LAKES

Location	Contaminant	Quantity in µg/kg	Date	Reference
<u>Lake Superior</u>				
Canadian Nearshore 28 sites	PCB	N.D.-250	1973	Kinkead and Chatterjee, 1974
	DDE	N.D.-7.1	1973	Kinkead and Chatterjee, 1974
	DDD	N.D.-2.7	1973	Kinkead and Chatterjee, 1974
	DDT	N.D.-1.5	1973	Kinkead and Chatterjee, 1974
	DEHP	0-1.5	1973	Kinkead and Chatterjee, 1974
Open Lake and Nearshore Areas	PCB	Trace-90	1974	Glooschenko <i>et al.</i> , 1976
	Dieldrin	N.D.-7	1974	Glooschenko <i>et al.</i> , 1976
	p,p'-DDE	N.D.-7	1974	Glooschenko <i>et al.</i> , 1976
	p,p'-TDE	N.D.-5	1974	Glooschenko <i>et al.</i> , 1976
	p,p'-DDT	N.D.-7	1974	Glooschenko <i>et al.</i> , 1976
	o,p'-DDT	N.D.	1974	Glooschenko <i>et al.</i> , 1976
	DDT	N.D.-12	1974	Glooschenko <i>et al.</i> , 1976
Nearshore, Western Area	PCB	7.0 ± 0.5	1975	Veith, <i>et al.</i> , 1977
Open Lake	PCB	N.D.-2.50	1973	IJC, 1977
	DDE	N.D.-4.2	1973	IJC, 1977
	DDD	N.D.-3.2	1973	IJC, 1977
	DDT	N.D.-1.3	1973	IJC, 1977
Open Lake	PCB	0.005-0.39	1978	Eisenreich <i>et al.</i> (1979)

TABLE 9. (CONT.)

Location	Contaminant	Quantity in $\mu\text{g}/\text{kg}$	Date	Reference	
<u>Lake Michigan</u>					
Southern Basin	$\Sigma\text{DDT}$	Surface sediments	18.5	1968	Torrey, 1976
		2-6 cm deep	6.3	1968	Torrey, 1976
		6-12 cm deep	3.4	1968	Torrey, 1976
	Dieldrin	Surface sediments	2.0	1968	Torrey, 1976
		2-12 cm deep	<0.5	1968	Torrey, 1976
Central Basin (11 stations)	$\Sigma\text{DDT}$	2.9	1973	Torrey, 1976	
	Dieldrin	0.1	1973	Torrey, 1976	
Central Basin (8 stations)	$\Sigma\text{DDT}$	14.4	1973	Torrey, 1976	
	Dieldrin	0.8	1973	Torrey, 1976	
Southern and Central Basins adjacent to WWTP outfalls)	Aroclor 1242	0.01-0.11	1974	Torrey, 1976	
	Aroclor 1254	0.01-0.03	1974	Torrey, 1976	
Southern and Central (19 stations)	Aroclor 1242	N.D.-0.08	1974	Torrey, 1976	
	Aroclor 1254	0.002-0.05	1974	Torrey, 1976	
Lake Wide Average	PCB	38.2	1978	PLUARG, 1978	
Waukegan Harbor	PCB as Aroclor 1248	5%	1976	Williams, 1976 in IJC, 1978	
	Pydraul 50E (triaryldrosphate PCB substitute)	0.8%	1975	Williams, 1976 in IJC, 1978	
Vicinity of Holland, Michigan	p,p'-DDT	5.6-7.1	1976	IJC, 1978	
Vicinity of White River	p,p'-DDT	7.4	1976	IJC, 1978	
Vicinity of Manistique River Harbor	PCB	600-17,500	1976	IJC, 1978	
Vicinity of Escanaba River Mouth	PCB	1600	1976	IJC, 1978	
Various Nearshore Areas	PCB	20-6,420	--	Klinert, 1976	

TABLE 9 (CONT.)

Location	Contaminant	Quantity in $\mu\text{g}/\text{kg}$	Date	Reference
<u>Lake Huron</u>				
Open Lake and Nearshore Areas	PCB	Trace-20	1974	Glooschenko, <u>et al.</u> , 1976
	Dieldrin	N.D.-trace	1974	Glooschenko, <u>et al.</u> , 1976
	p,p'-DDE	N.D.-10	1974	Glooschenko, <u>et al.</u> , 1976
	p,p'-TDE	N.D.-9	1974	Glooschenko, <u>et al.</u> , 1976
	p,p'-DDT	N.D.-12	1974	Glooschenko, <u>et al.</u> , 1976
	o,p'-DDT	N.D.-1	1974	Glooschenko, <u>et al.</u> , 1976
	$\Sigma$ DDT	N.D.-22	1974	Glooschenko, <u>et al.</u> , 1976
Saginaw Bay	DBP	<200-290	1974-75	IJC, 1977
	PCB	N.D.-853	1974-75	IJC, 1977
	DDE	N.D.-13.6	1974-75	IJC, 1977
	Dieldrin	N.D.-4.5	1974-75	IJC, 1977
	DDD	N.D.-14.3	1974-75	IJC, 1977
	p,p'-DDT	N.D.-6.7	1974-75	IJC, 1977
	o,p'-DDT	N.D.-7	1974-75	IJC, 1977
<u>Lake Erie</u>				
Total Lake	PCB	4-800	1971	IJC, 1978
	DDE	0.2-136	1971	IJC, 1978
	TDE	0.3-186	1971	IJC, 1978
	Dieldrin	0.5-5.0	1971	IJC, 1978
Non-Depositional Zone	PCB	8-800	1971	IJC, 1978
	DDE	0.2-22.7	1971	IJC, 1978
	TDE	0.3-146	1971	IJC, 1978
	Dieldrin	0.5-3.5	1971	IJC, 1978
Total Basin	PCB	4-660	1971	IJC, 1978
	DDE	0.5-136	1971	IJC, 1978
	TDE	0.4-186	1971	IJC, 1978
	Dieldrin	0.5-5.0	1971	IJC, 1978
Western Basin	PCB	4-660	1971	IJC, 1978
	DDE	1.7-136	1971	IJC, 1978
	TDE	2.8-186	1971	IJC, 1978
	Dieldrin	0.6-3.3	1971	IJC, 1978

TABLE 9 (CONT.)

Location	Contaminant	Quantity in µg/kg	Date	Reference
<u>Lake Erie (Cont.)</u>				
Central Basin	PCB	12-330	1971	IJC, 1978
	DDE	0.5-18.7	1971	IJC, 1978
	TDE	0.4-55	1971	IJC, 1978
	Dieldrin	0.5-5.0	1971	IJC, 1978
Eastern Basin	PCB	12-320	1971	IJC, 1978
	DDE	0.6-30	1971	IJC, 1978
	TDE	0.5-61	1971	IJC, 1978
	Dieldrin	0.6-3.8	1971	IJC, 1978
Nearshore Lake Erie	DDE	0.002-0.018	1973	IJC, 1978
	TDE	0.002-0.030	1973	IJC, 1978
	o,p'-DDT	0.002-0.003	1973	IJC, 1978
	p,p'-DDT	0.006-0.015	1973	IJC, 1978
	PCB	0.08-0.38	1973	IJC, 1978
	DEHP	1.0-5.0	1973	IJC, 1978
	DBP	3.0-6.0	1973	IJC, 1978
<u>Lake Ontario</u>				
Midlake Ontario	DDE	11.0	1975	IJC, 1977
	DDD	5.4	1975	IJC, 1977
	DDT	2.8	1975	IJC, 1977
	Σ DDT	19.0	1975	IJC, 1977
	Dieldrin	0.5	1975	IJC, 1977
	PCB	79	1975	IJC, 1977
Open Lake, Eastern Basin	DDE	16	1975	IJC, 1977
	DDD	31	1975	IJC, 1977
	DDT	7.4	1975	IJC, 1977
	Σ DDT	54	1975	IJC, 1977
	Dieldrin	2.1	1975	IJC, 1977
	PCB	N.D.	1975	IJC, 1977
Various Nearshore	DDE	4.8-12	1975	IJC, 1977
	DDD	1.5-15	1975	IJC, 1977
	DDT	0.2-12	1975	IJC, 1977
	Σ DDT	10-39	1975	IJC, 1977
	Dieldrin	0.5-2.6	1975	IJC, 1977
	PCB	43-245	1975	IJC, 1977
Cobourg Harbor	Lindane	N.D.	--	IJC, 1978
	Heptachlor	N.D.	--	IJC, 1978
	Aldrin	N.D.	--	IJC, 1978

TABLE 9 (CONT.)

Location	Contaminant	Quantity in µg/kg	Date	Reference
<u>Lake Ontario (Cont.)</u>				
Cobourg Harbor (Cont.)	Heptachlor Epoxide	N.D.	--	IJC, 1978
	p,p'-DDE	N.D.	--	IJC, 1978
	Dieldrin	N.D.-1.0	--	IJC, 1978
	p,p'-DDD	N.D.-2.0	--	IJC, 1978
	p,p'-DDT	N.D.-1.0	--	IJC, 1978
	p,p'-DDT	N.D.	--	IJC, 1978
	Endrin	N.D.	--	IJC, 1978
	Cis/trans chlordane	N.D.	--	IJC, 1978
	α Endosulfan	N.D.-1.0	--	IJC, 1978
	β Endosulfan	N.D.	--	IJC, 1978
	Meltoxychlor	N.D.	--	IJC, 1978
	PCB	100-500	--	IJC, 1978
St. Lawrence River Drainage Basin Unspecified	PCB	10.0	1971	Dennis, 1976
	PCB	2.0-800	1972	Dennis, 1976
	PCB	5.0-13,000	1973	Dennis, 1976
	PCB	3.0-700	1974	Dennis, 1976

As a result of these phenomena, and because of the relative ease of quantifying organochlorine compounds at the mg/kg as compared with levels in the parts per billion or trillion range, Great Lakes fish have been extensively utilized as both monitoring and indicator organisms. As was mentioned for other sections, an exhaustive analysis of extant data on Great Lakes fish is beyond the scope of this effort, but a synopsis of major findings is provided in Tables 10 and 11.

For additional summaries of large data sets on organochlorine contaminants in Great Lakes fish, the reader is referred to Upper Lakes Reference Group, I.J.C., (1977); International Joint Commission (1977); and International Joint Commission (1978).

### Adult Human Exposure

It is becoming increasingly evident that for the first time in more than fifty years of Great Lakes research, it is necessary to consider man as both an interactive and reciprocal part of the Great Lakes ecosystem. Prior to this time, the human species has had the luxury of regarding itself as simply a source or input term. Because of the biomagnification phenomenon in the Great Lakes ecosystem, man too has become a reservoir or sink in the ecosystem organochlorine compound mass conservation expression as a result of his consumption of Great Lakes fish.

Public exposure to orally ingested PCBs is rather effectively controlled through the activities of the United States Food and Drug Administration's tolerance levels set for these substances. Established in 1973, the FDA levels restrict fish, shellfish, and the fat portions of poultry above 5.0 parts per million (mg/kg); require levels no greater than 2.5 parts per million (mg/kg fat portion) in milk and dairy products; 0.5 parts per million (mg/kg) in eggs; 0.2 parts per million (mg/kg) in infant and junior foods; and limit PCBs in packaging materials to 10 parts per million (mg/kg). At the time of the preparation of this summary, FDA has undertaken steps to reduce the tolerance levels/ permissible amounts to 2.0 parts per million (mg/kg) in fish and shellfish; 1.5 parts per million (mg/kg) in milk and milk products; 0.3 parts per million (mg/kg) in eggs; and 3.0 parts per million (mg/kg) in poultry. Establishment of these reductions in acceptable values has been delayed in part by action enjoining the activities of the agency. The lower levels for eggs, milk and dairy products, and poultry went into effect on August 28, 1979, but the National Fisheries Institute filed objections to the proposed standards for fish and requested a hearing.

The theoretical effects of adopting and implementing regulatory standards of this sort are two:

1. Reduction of the absolute magnitude of exposure per unit of time, and hence measurable levels or body burdens among the general population, and

TABLE 10. POLYCHLORINATED BIPHENYLS OBSERVED IN FISH OF THE GREAT LAKES

Location	Species	Date	Quantity in mg/kg	Reference
<u>Lake Superior</u>				
Nearshore, Ontario	Lake Trout	1974	0.387-1.796	Upper Lakes Ref. Group, 1977
	Walleye	1974	0.243-3.635	Upper Lakes Ref. Group, 1977
	Whitefish	1974	0.125-3.635	Upper Lakes Ref. Group, 1977
	Herring	1974	0.193-0.757	Upper Lakes Ref. Group, 1977
Nearshore, Minnesota	Mottled Sculpin	1974	0.035-0.600	Upper Lakes Ref. Group, 1977
Nearshore, Michigan	Lake Trout	1974	N.D.-0.46	Upper Lakes Ref. Group, 1977
	Siscowett Trout	1974	1.95-8.37	Upper Lakes Ref. Group, 1977
	Whitefish	1974	0.27-0.89	Upper Lakes Ref. Group, 1977
	Herring	1974	0.22-1.03	Upper Lakes Ref. Group, 1977
	Mottled Sculpin	1974	N.D.-0.46	Upper Lakes Ref. Group, 1977
Nearshore, Wisconsin	Bullhead	1974	0.017-0.050	Upper Lakes Ref. Group, 1977
	Northern Pike	1974	0.020-0.030	Upper Lakes Ref. Group, 1977
	Walleye	1974	0.080-0.218	Upper Lakes Ref. Group, 1977
	Yellow Perch	1974	0.046-0.05	Upper Lakes Ref. Group, 1977
	White Sucker	1974	0.024	Upper Lakes Ref. Group, 1977
	Rainbow Smelt	1974	0.202-0.263	Upper Lakes Ref. Group, 1977
	Brown Trout	1974	0.145	Upper Lakes Ref. Group, 1977
	Rainbow Trout	1974	0.065-0.110	Upper Lakes Ref. Group, 1977
Open Lake	Slimy Sculpin	1974	0.16-0.36	Upper Lakes Ref. Group, 1977
	Burbot	1974	1.40-1.59	Upper Lakes Ref. Group, 1977

TABLE 10. (CONT.)

Location	Species	Date	Quantity in mg/kg	Reference
<u>Lake Superior (Cont.)</u>				
Open Lake (Cont.)	Lake Trout	1974	0.80-4.32	Upper Lakes Ref. Group, 1977
Nearshore, Apostle Islands	Lake Trout (Gillets- Whole Fish)	1974	1.68-1.80	Upper Lakes Ref. Group, 1977
Nearshore, Wisconsin	Rainbow Smelt	1974-1976	0.250-0.270	IJC, 1978
	Rainbow Trout	1974-1976	0.40-0.110	IJC, 1978
	Brown Trout	1974-1976	0.130-0.235	IJC, 1978
	Perch	1974-1976	0.02-0.450	IJC, 1978
	Northern Pike	1974-1976	0.03-0.65	IJC, 1978
	Bullhead	1974-1976	0.003-0.03	IJC, 1978
	Carp	1974-1976	0.19-1.34	IJC, 1978
Isle Royale Area	Siscowett Trout	1975	2.06-6.90	IJC, 1978
	Whitefish	1974	0.72-3.77	IJC, 1978
Nearshore, Michigan	Lake Trout	1975	0.62-2.58	IJC, 1978
	Siscowett Trout	1975	2.06-31.20	IJC, 1978
	Herring	1974	0.22-1.03	IJC, 1978
	Chubs	1974	0.62-0.69	IJC, 1978
Nearshore, Wisconsin	Lake Trout	1974	2.7	U.S. Fish and Wildlife Ser., 1974
	Lake Whitefish	1974	0.7	U.S. Fish and Wildlife Ser., 1974
	Bloater	1974	1.2-2.7	U.S. Fish and Wildlife Ser., 1974
Nearshore, Michigan	Lake Trout	1974	3.7-4.5	U.S. Fish and Wildlife Ser., 1974
	Herring	1974	1.1	U.S. Fish and Wildlife Ser., 1974
	Bloater	1974	0.61-0.79	U.S. Fish and Wildlife Ser., 1974
Isle Royale	Lake Trout	1974-1976	0.3-1.72	Swain, 1978
	Siscowett Trout	1974-1976	5.4-8.4	Swain, 1978
	Whitefish	1974-1976	0.37	Swain, 1978

TABLE 10. (CONT.)

Location	Species	Date	Quantity in mg/kg	Reference
<u>Lake Michigan</u>				
	Bloater	1972	3.12-8.17	Willford, et al., 1976
	Bloater	1973	4.20-6.33	Willford, et al., 1976
	Bloater	1974	4.83-6.22	Willford, et al., 1976
	Coho Salmon	1972	4.93-15.4	Willford, et al., 1976
	Coho Salmon	1973	8.24-16.6	Willford, et al., 1976
	Coho Salmon	1974	6.99-16.3	Willford, et al., 1976
	Lake Trout	1972	3.53-25.3	Willford, et al., 1976
	Lake Trout	1973	9.36-30.6	Willford, et al., 1976
	Lake Trout	1974	7.05-47.4	Willford, et al., 1976
Vicinity Saugatuck and East-Central Lake Area (annual mean values)	Bloater	1972	5.66	U.S. Fish and Wildlife Ser. in IJC, 1978
	Bloater	1973	5.24	U.S. Fish and Wildlife Ser. in IJC, 1978
	Bloater	1974	5.57	U.S. Fish and Wildlife Ser. in IJC, 1978
	Bloater	1975	4.54	U.S. Fish and Wildlife Ser. in IJC, 1978
	Bloater	1976	4.11	U.S. Fish and Wildlife Ser. in IJC, 1978
	Coho Salmon	1972	10.93	U.S. Fish and Wildlife Ser. in IJC, 1978
	Coho Salmon	1973	12.17	U.S. Fish and Wildlife Ser. in IJC, 1978
	Coho Salmon	1974	10.45	U.S. Fish and Wildlife Ser. in IJC, 1978
	Coho Salmon	1975	10.77	U.S. Fish and Wildlife Ser. in IJC, 1978

TABLE 10. (CONT.)

Location	Species	Date	Quantity in mg/kg	Reference
<u>Lake Michigan (Cont.)</u>				
Vicinity Saugatuck and East-Central Lake Area (Cont.)	Coho Salmon	1976	9.21	U.S. Fish and Wildlife Ser. in IJC, 1978
	Lake Trout	1972	12.86	U.S. Fish and Wildlife Ser. in IJC, 1978
	Lake Trout	1973	18.93	U.S. Fish and Wildlife Ser. in IJC, 1978
	Lake Trout	1974	22.91	U.S. Fish and Wildlife Ser. in IJC, 1978
	Lake Trout	1975	22.28	U.S. Fish and Wildlife Ser. in IJC, 1978
	Lake Trout	1976	18.68	U.S. Fish and Wildlife Ser. in IJC, 1978
Nearshore, Michigan	Salmon	1974	0.22-1.64	Mich. Dept. Agr. in IJC, 1978
	Lake Trout	1974-1975	0.99-27.80	Mich. Dept. Agr. in IJC, 1978
	Chub	1974	2.87-3.74	Mich. Dept. Agr. in IJC, 1978
Nearshore, Wisconsin	Carp	1976	16.3	Kleinert, 1976
	Lake Trout	1976	2.9-33.8	Kleinert, 1976
<u>Lake Huron</u>				
Vicinity of Bay Port, Michigan	Yellow Perch	1974	2.2-3.9	U.S. Fish and Wildlife Ser. in IJC, 1978
	Carp	1971	3.0-4.7	U.S. Fish and Wildlife Serv. in IJC, 1978
Vicinity of Alpena, Michigan	Yellow Perch	1974	0.65-0.94	U.S. Fish and Wildlife Serv. in IJC, 1978
	White Sucker	1974	0.73	U.S. Fish and Wildlife Ser. in IJC, 1978

TABLE 10. (CONT.)

Location	Species	Date	Quantity in mg/kg	Reference
<u>Lake Huron (Cont.)</u>				
Vicinity of Alpena, Michigan (Cont.)	Whitefish	1974	0.46	U.S. Fish and Wildlife Ser. in IJC, 1978
Nearshore, Michigan	Brown Trout	1974	0.35-1.93	Mich. Dept. Agr. in IJC, 1978
	Carp	1974	1.06-4.30	Mich. Dept. Agr. in IJC, 1978
	Chinook Salmon	1974	1.29-2.97	Mich. Dept. Agr. in IJC, 1978
	Yellow Perch	1974	0.02-0.23	Mich. Dept. Agr. in IJC, 1978
	Brown Trout	1975	0.64-1.70	Mich. Dept. Agr. in IJC, 1978
	Carp	1975	0.92-2.75	Mich. Dept. Agr. in IJC, 1978
	Lake Trout	1975	0.99-5.7	Mich. Dept. Agr. in IJC, 1978
	Rainbow Smelt	1975	0.69-0.80	Mich. Dept. Agr. in IJC, 1978
	Whitefish	1975	0.20-0.43	Mich. Dept. Agr. in IJC, 1978
Nearshore, Michigan	Brown Trout	1974-1975	1.1-1.3	Upper Lakes Ref. Group, 1977
	Perch	1974-1975	N.D.-0.20	Upper Lakes Ref. Group, 1977
	Rainbow Trout	1974-1975	0.94	Upper Lakes Ref. Group, 1977
	Whitefish	1974-1975	N.D.-0.34	Upper Lakes Ref. Group, 1977
	Chinook	1974-1975	2.31	Upper Lakes Ref. Group, 1977
Nearshore, Ontario	Perch	1974-1975	0.003-0.40	Upper Lakes Ref. Group, 1977
	White Sucker	1974-1975	0.125-1.153	Upper Lakes Ref. Group, 1977
	Rainbow Trout	1974-1975	0.09-2.17	Upper Lakes Ref. Group, 1977
	Walleye	1974-1975	0.258-0.827	Upper Lakes Ref. Group, 1977
	Northern Pike	1974-1975	0.024-0.085	Upper Lakes Ref. Group, 1977

TABLE 10. (CONT.)

Location	Species	Date	Quantity in mg/kg	Reference
<u>Lake Huron (Cont.)</u>				
Open Lake	Burbot	1974-1975	0.77-2.35	Upper Lakes Ref. Group, 1977
	Bloater Chubs	1974-1975	0.89-361	Upper Lakes Ref. Group, 1977
	Slimy Sculpins	1974-1975	0.52-0.77	Upper Lakes Ref. Group, 1977
<u>Lake Erie</u>				
Nearshore	Shad	1977	0.69-1.72	Herdendorf, <u>et</u> <u>al.</u> , 1978
	Perch	1977	0.35-1.34	Herdendorf, <u>et</u> <u>al.</u> , 1978
	Catfish	1977	3.14-3.85	Herdendorf, <u>et</u> <u>al.</u> , 1978
	Drum	1977	0.26-0.36	Herdendorf, <u>et</u> <u>al.</u> , 1978
Nearshore, Michigan	Carp	1974	3.7-3.9	Mich. DNR in IJC, 1978
	Catfish	1974	2.97-3.0	Mich. DNR in IJC, 1978
	Drum	1974	0.52	Mich. DNR in IJC, 1978
	Yellow Perch	1974	0.03	Mich. DNR in IJC, 1978
	Walleye	1974	0.22	Mich. DNR in IJC, 1978
	White Bass	1974	2.18	Mich. DNR in IJC, 1978
	Carp	1975	3.17-3.9	Mich. DNR in IJC, 1978
	Catfish	1975	3.29-5.65	Mich. DNR in IJC, 1978
	Drum	1975	0.11	Mich. DNR in IJC, 1978
	Salmon	1975	0.38	Mich. DNR in IJC, 1978
	Walleye	1975	0.34	Mich. DNR in IJC, 1978
	White Bass	1975	0.55-1.78	Mich. DNR in IJC, 1978
	Rainbow Trout	1975	0.70	Mich. DNR in IJC, 1978

TABLE 10. (CONT.)

Location	Species	Date	Quantity in mg/kg	Reference
<u>Lake Erie (Cont.)</u>				
Nearshore, Michigan (Cont.)	Chinook Salmon	1975	0.25	Mich. DNR in IJC, 1978
<u>Lake Ontario</u>				
Nearshore	American Eel	1976	0.12-14.77	IJC, 1977
	Largemouth Bass	1976	0.09-2.92	IJC, 1977
	Smallmouth Bass	1976	0.45-10.93	IJC, 1977
	White Bass	1976	0.83-5.17	IJC, 1977
	Black Crappie	1977	0.51-0.89	IJC, 1977
	Brown Bullhead	1977	0.30-2.81	IJC, 1977
	Muskellunge	1977	1.93-4.05	IJC, 1977
	Northern Pike	1977	0.58-4.17	IJC, 1977
	Yellow Perch	1977	0.22-1.72	IJC, 1977
	White Perch	1977	0.86-9.70	IJC, 1977
	Coho Salmon	1977	1.60-10.27	IJC, 1977
	Chinook Salmon	1977	2.36-13.47	IJC, 1977
	Brown Trout	1977	1.31-15.14	IJC, 1977
	Lake Trout	1977	1.38-18.30	IJC, 1977
	Rainbow/Steelhead	1977	10.64	IJC, 1977
	Walleye	1977	0.08-1.21	IJC, 1977
St. Lawrence River (edible portions of fish)	Smallmouth Bass	1976	0.25-16.17	IJC, 1977
	Northern Pike	1976	1.18-3.93	IJC, 1977
	Yellow Perch	1976	0.46-4.26	IJC, 1977
	White Perch	1976	0.66-11.80	IJC, 1977
	Walleye	1976	1.41-6.52	IJC, 1977
Eastern Nearshore Area (edible portions of fish)	Catfish	1976	7.9-12.42	IJC, 1977
	Pike	1976	0.22-0.89	IJC, 1977
	Coho Salmon	1976	2.0-5.38	IJC, 1977
	Sucker	1976	0.96-1.15	IJC, 1977
	Carp	1976	1.3-3.29	IJC, 1977
	White Perch	1976	0.82-2.8	IJC, 1977
	American Eel	1976	4.4-8.5	IJC, 1977
	Yellow Perch	1976	0.28-0.52	IJC, 1977
	Bullhead	1976	0.21-0.41	IJC, 1977
	Rainbow Smelt	1976	1.12-1.22	IJC, 1977
	Sheepshead	1976	0.89-6.6	IJC, 1977
	Rock Bass	1976	0.11-5.21	IJC, 1977
Adjacent to Credit River	Coho Salmon	1975	2.1-22.5	IJC, 1977

TABLE 10. (CONT.)

Location	Species	Date	Quantity in mg/kg	Reference
<u>Lake Ontario (Cont.)</u>				
Various Nearshore Areas	Alewife	1975	0.14-381	Haile, <u>et al.</u> , 1975
	Smelt	1975	1.40-3.49	Haile, <u>et al.</u> , 1975
	Slimy Sculpin	1975	1.58-9.17	Haile, <u>et al.</u> , 1975
	Rock Bass	1976	N.D.-2.60	IJC, 1978
	White Perch	1975	4.5-6.4	IJC, 1978
	Yellow Perch	1975	1.7-3.3	IJC, 1978
	Rock Bass	1975	2.0-2.4	IJC, 1978

TABLE 11  
 REPRESENTATIVE MEAN CONTAMINANT RANGES IN SELECTED  
 GREAT LAKES FISH

<u>LAKE</u>	<u>SPECIES OF FISH</u>	<u><math>\Sigma</math>DDT (mg/kg)</u>	<u>PCB (mg/kg)</u>
SUPERIOR	Lake Trout	0.5-1.0	2.0-4.0
	Siscowet Trout	5.0-10.0	8.0-15.0
	Yellow Perch	0.01-0.05	0.02-0.05
MICHIGAN	Lake Trout	5.0-10.0	18.0-20.0
	Coho Salmon	3.0-10.0	7.0-17.0
	Yellow Perch	-	0.5-1.0
HURON	Lake Trout	2.0-4.0	3.0-5.0
	Yellow Perch	0.04-0.4	0.04-0.4
ERIE	Coho Salmon	0.3-0.5	0.5-1.5
	Yellow Perch	0.1-0.5	0.03-0.1
ONTARIO	Lake Trout	2.0-4.0	6.0-8.0
	Coho Salmon	1.0-2.0	6.0-8.0
	Yellow Perch	0.03-0.1	0.5-3.0

2. A tendency toward a leveling effect in exposure potentials throughout the population-at-large, leading to a more nearly uniform exposure experience across a population.

While exposure among the general population via food sources has been demonstrated, particularly prior to the 1973 limitations imposed by the Food and Drug Administration (Kolbye, 1972; Biros *et al.*, 1970; Finklea *et al.*, 1972), there appears to be no data from "market basket" studies supporting substantial differences in current levels of exposure from food sources between one area of the country and another. Jelinek and Corneliussen (1976) report that "the occurrence of PCBs has narrowed to the point where freshwater fish are now the primary source of PCBs in our diet." They further note that "the daily intake of PCB for the average citizen is low, since his consumption of freshwater fish is low." They also add, however, "PCB intake could be quite different for those people whose diets include substantial quantities of sports fish." Thus, if the exposure potential from food sources for the general population can be assumed to be approaching a uniform level, and, as a result of FDA limitation, that level is at least an order of magnitude below the exposure potential for consumption of Great Lakes fish, an argument for consideration of the health consequences of Great Lakes Basin residents is raised.

In addition to the potential for human exposure from consumption of Great Lakes fish, Kutz and Strassman (1976) indicate three primary routes of intake for humans: 1) ingestion, 2) respiration, and 3) absorption through skin and mucous membranes. Thus, for the bulk of the population at risk, beyond food sources, additional routine inputs among non-occupationally exposed individuals are two: potable drinking water and respired ambient air. Table 12 considers the anticipated annual individual exposure from each of these sources, and the combined anticipated exposure from both sources considered together. While the calculation does not represent exposure experience, particularly for the residents of highly industrialized urban population centers, it does suggest a useful order of magnitude comparison relative to additional sources of ingested organochlorine contaminants.

On the basis of this calculation, and a comparison of the data in Tables 10 and 11, it is theoretically possible in terms of exposure potential to breathe the air in the Lake Michigan Basin and drink its polished drinking water for a period exceeding a lifetime before realizing the same effective exposure received from consuming a single one pound fish meal of Lake Michigan lake trout.

Public consumption of Great Lakes fish heavily contaminated with organochlorine compounds, including PCB, is effectively controlled through embargo of commercial catches in the United States by the Food and Drug Administration. However, substantial and significant exposure to these species is possible for sports fishermen and their families.

A preliminary study by the Michigan Department of Public Health (Humphrey, undated) has shown that in the 18 counties of the State of Michigan, which border and abut on Lake Michigan, there are 381,600

TABLE 12. CALCULATED PCB EXPOSURE POTENTIAL FOR NON-URBAN GREAT LAKES BASIN RESIDENTS FROM DRINKING WATER AND RESPIRATION

ORAL EXPOSURE TO PCB FROM GREAT LAKES DRINKING WATER

Assumptions:	Mean raw water value for PCB in Great Lakes waters	4 ng/l
	Finished drinking water PCB values are no higher than raw water values	
	Mean daily water intake per person	2
	Total Daily Oral Exposure Rate From Drinking Water	8 ng/
	Total Annual Oral Exposure Rate From Drinking Water	2.9 µg/yr.

RESPIRATORY PCB EXPOSURE FROM AMBIENT AIR LEVELS

Assumptions:	Mean level of ambient PCB in in the atmosphere	1.5 ng/m
	Complete and instantaneous sorption and selective up- take of PCB at Alveolar sur- faces in the lung	
	Average rate of respiration	20 respiration/min 1,200 respiration/hr. 28,800 respiration/day
	Mean tidal volume of lungs	≈500 ml
	Total Air Respired Per Day	14,400 l/day (14.4 m <sup>3</sup> /day)
	Average Daily Respiratory Exposure Rate	14.4x1.5 = 21.6 ng/day
	Annual Exposure Rate	7.8 µg/yr.

TOTAL ANTICIPATED COMBINED EXPOSURES FROM AIR AND WATER

2.9 µg/yr. + 7.8 µg/yr. = 10.7 µg/yr.

licensed sports fishermen who take, and presumably consume 13,975,650 pounds of salmonid fishes from Lake Michigan each year. The vast majority of these species are Lake Trout, Coho and Chinook Salmon, particularly of the size range which is significantly contaminated with PCB and other organochlorine compounds. Statistically, each of these fishermen would then consume approximately three-quarters of a pound of fish per week or 36 pounds per annum. This value is over three times the national average for consumption of fish from commercial sources (11.8 lbs/capita/yr), and substantially above the recommended maximum for Lake Michigan fish set by the Michigan State Department of Public Health (24 pounds/capita/yr).

Humphrey and his co-workers (Humphrey, undated) surveyed a population of 161 consumers of Lake Michigan fish. Over two fishing seasons in the study, these workers found that the most frequently consumed quantity of fish was in the 24-35 pounds per year range (Figure 2). Consumption above this mean value was found, however. The highest reported ingestion over the two year period was 180 pounds per year, while the maximum single season consumption was 260 pounds per year.

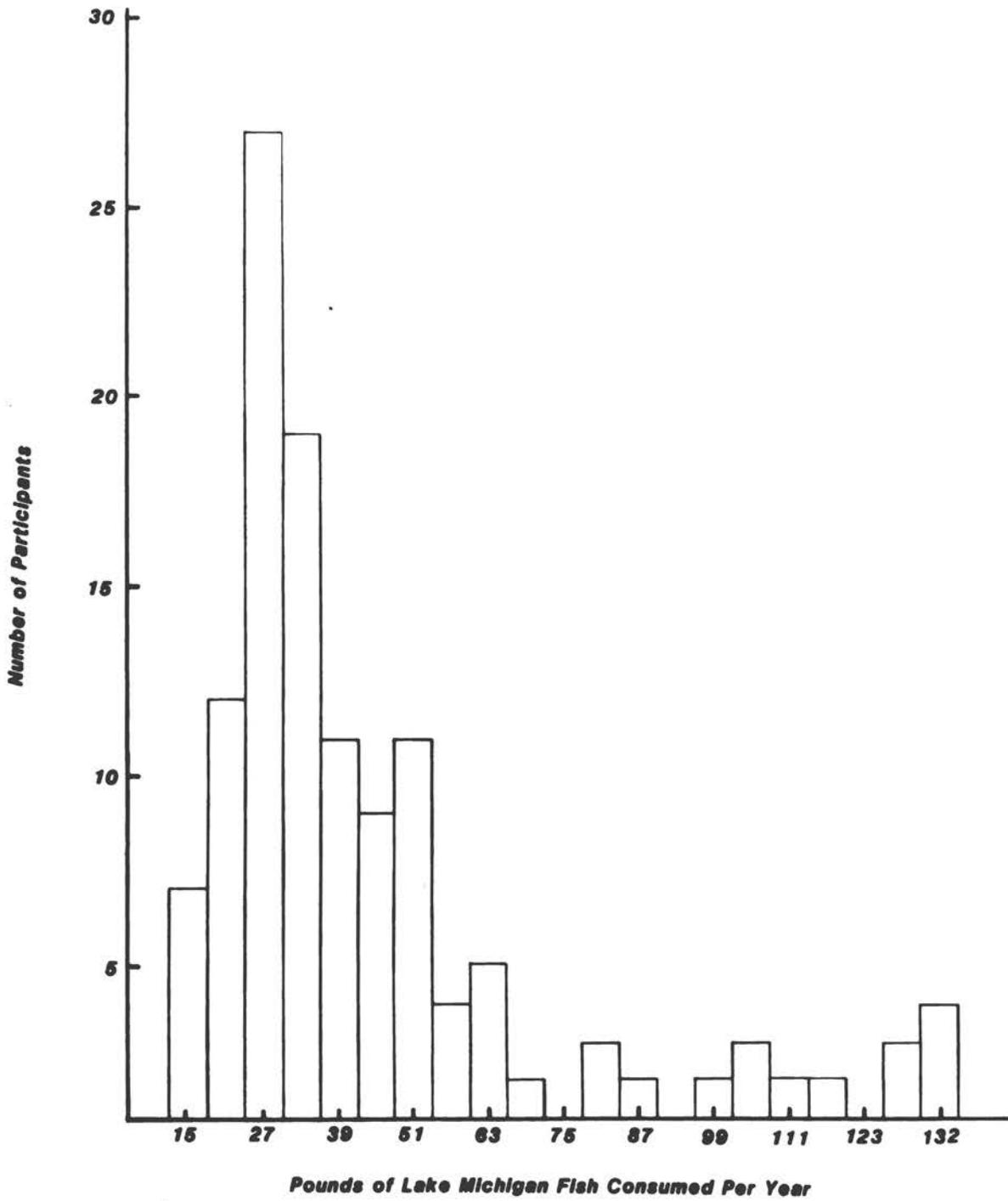
#### Effects of Acute Exposure

The human health effects from acute PCB exposure is poorly understood. To date, information on the impacts of PCB exposure has been chiefly confined to acute effects from industrial exposure and a single, although extensive, episode in Japan in which a disease called Yusho ("oil disease") resulted from consumption of rice oil contaminated with a commercial PCB formulation called Kanechlor 400 (Kuratsune et al., 1972).

Yusho is considered as an acute or sub-acute poisoning with PCB. General symptomatology includes retarded growth; neuroendocrine disturbances; respiratory disorders; abnormal lipid metabolism; increased pigmentation of skin, nail beds, conjunctiva, and gingiva; swelling and hypersecretion of the Meibomian gland accompanied by palpebral edema; and cutaneomucosal lesions, including suppurative chloracne (Kuratsune et al., 1976; Higuchi (editor), 1976).

Koda and Masuda (1975) examined 72 patients, the majority of whom were still suffering the effects of chloracne, nail deformation, and Meibomian hypersecretion five years after the initial ingestion of the contaminated rice oil. The inability of body systems to effectively metabolize these substances is demonstrated by this contamination of symptomatology.

Although reports of the toxic symptoms of Yusho have been attributed to PCB intoxication, the suspected occurrence of other toxic substances in the Japanese PCB preparations has called for inquiry into the composition of the contaminated rice oil (Nagayama et al., 1977). Kuratsune et al. (1976) report that Nagayama et al. (1975) examined the original contaminated rice oil from three Yusho positive families for polychlorinated dibenzo-p-dioxins (PCDDs) and for polychlorinated dibenzofurans (PCDFs). While the former was not observed, the latter was found in all cases, the highest concentration observed approaching 5.0 mg/l. The major constituents of the observed peaks were tetra and penta-chlorodibenzofurans, one of which was apparently 2, 3, 7, 8 -



**FIGURE 2. FREQUENCY DISTRIBUTION OF REPORTED GREAT LAKES FISH CONSUMPTION, EXPOSED POPULATION**

*(from Humphrey, undated)*

tetrachlorodibenzofuran, reported to be among the most toxic of this chemical species (Stalling, 1980). Even with these data, however, the question of the etiologic agent or agents of Yusho symptomatology is not fully resolved.

To date in the Great Lakes basin, there appears to be only a single published report of an acute episode of classic Yusho-like symptomatology (Ingersoll, 1977). This report, designed for consumption by the general public, regrettably lacks the precise documentation usually associated with a medical case history. It does, however, note gross symptomatology usually attributed to the PCB intoxication associated with Yusho. Further, it specifies a direct link between the onset of symptoms and the consumption of Lake Michigan fish which had been embargoed as a result of excessive contamination with organochlorine compounds.

Additional cases of Yusho-like symptoms have been noted in the Great Lakes basin, usually among persons for whom Great Lakes fish constitute the principal protein source, but subsequent examination has failed to demonstrate the elevated blood levels of PCB usually associated with Yusho poisoning (Weininger, 1980).

#### Effects of Chronic Exposure

The ultimate cumulative human health effects of long term low level exposure to PCB are even less well understood than the acute impacts on the human population. Intermediate consequences of exposure, however, appear to be quantifiable.

#### Accumulation in Adipose Tissue

Because PCB compounds are low in biodegradability and poorly metabolized, they tend to accumulate and be retained in biologic tissues consisting of, or containing quantities of lipids and oils (Allen *et al.*, 1974; Kimbrough *et al.*, 1973; Kimbrough, 1976). Several authors have reported results from samples of adipose tissue taken at random from the U.S. population. Price and Welch (1972) note that 41 to 45 percent of the population studied had levels of 1.0 mg/kg PCB or greater in their adipose tissue. Yobs (1972) found 31.1 percent of 637 samples collected during 1971 at a level of 1 mg/kg PCB or greater. Kutz and Strassman (1976) report 35.1 percent above 1 mg/kg for the 1277 samples analyzed for 1973, and 40.3 percent above 1 mg/kg for the 1047 individuals studied in 1974.

Among Japanese populations exposed to Kanechlor 400, Kuratsune *et al.* (1976) report levels of 1.2-1.4 mg/kg on a whole basis in adipose tissue in patients with Yusho, as compared with 0.4-1.0 among unexposed individuals. When these data were calculated on a fat basis, the Yusho patients were observed to fall in a range of 2.1-8.5 mg/kg and contrasted with 0.7-1.4 mg/kg among unexposed persons.

Grant *et al.* (1976) reviewing data from a national survey by Health and Welfare Canada note differences in PCB residues in adipose tissue among the various provinces and regions of Canada. While all samples of adipose tissue examined contained detectable levels of PCB, in Ontario

Province, 49 percent of the population surveyed had levels at greater than 1 mg/kg. Quebec Province, the Atlantic and Western regions were similar to each other, but markedly reduced from the Ontario values with 22 percent, 25 percent and 22 percent of the samples above 1 mg/kg, respectively. The central region (Manitoba and Saskatchewan) was significantly lower with only 9 percent of the population surveyed above 1 mg/kg. Table 13 not only shows this trend among the mean distributions of PCB residues observed, but displays substantial differences in values between the sexes.

Grant *et al.* (1976) reporting the work of Holdrient *et al.* (1975) note a range from non-detectable levels to 18 mg/kg PCB on a fat basis with a mean of 2.5 mg/kg for 282 adipose samples from Ontario Province residents in 1971-72. 1973-74 data showed a range of 0.6 to 11.0 mg/kg with a mean value of 2.3 mg/kg for 129 similar samples. The work of Mes *et al.* (1977) provides more recent data on PCBs, as well as a variety of other organochlorine compounds in adipose tissue (Table 14).

#### Accumulation in Blood

Another useful intermediate index of human exposure is the accumulation of organochlorine compounds, including PCB, in human blood. That exposure is linked to accumulations in circulating blood titer, is adequately demonstrated by Kuratsune *et al.* (1976) reporting Japanese data on the acute Yusho exposure (Table 15). These workers note that single acute exposure Yusho patients have significantly higher blood burdens than do persons who were not exposed to orally ingested Kanechlor 400, but both groups are substantially lower in circulating blood titer than workers occupationally exposed during the production of PCBs. While differing in absolute magnitude, similar findings are reported among three identical groups by Kuwabara *et al.* (1978).

Of particular interest is the work of Kuwabara *et al.* (1979), who demonstrated a rapid uptake of PCB by two human subjects who ingested fish meals containing 128 and 181  $\mu\text{g}$  PCB, respectively. In both cases, circulating blood titers of PCB rose dramatically within a few hours. Maximum values were achieved in 5 hours in the subject exposed to 128  $\mu\text{g}$  PCB, and in 3 hours for the individual who was administered 181  $\mu\text{g}$ . Corresponding control experiments showed little or no increase through time.

#### Intermediate Effects of Consumption of Great Lakes Fish

As was previously noted, sport fishing in the Great Lakes represents an uncontrolled source of PCB contaminated fish for human consumption. Willford (1980) has reported PCB values for 20-28 inch Lake Michigan Lake Trout in a range of 12.86 to 22.91 mg/kg for the years 1972 through 1974, and levels of PCB in 20 to 32 inch Coho Salmon of between 10.93 and 12.17 for the same period.

Given the levels and the sizable proportion of fish taken and presumably consumed, concern was developed by the Michigan State Health Department for the health impacts of PCB exposure from Great Lakes fish

TABLE 13

PCB RESIDUES (mg/kg) IN HUMAN ADIPOSE TISSUE<sup>1</sup>

<u>Sex</u>	<u>Atlantic</u>	<u>Quebec</u>	<u>Ontario</u>	<u>Central</u>	<u>Western</u>	<u>Canada</u>
M	0.758	1.125	1.165	0.6211	0.977	1.020
F	0.593	0.723	0.859	0.377	0.684	0.685
M & F	0.727	0.969	1.070	0.499	0.898	0.907

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<sup>1</sup>From Grant et al. (1976)

TABLE 14  
 CHLORINATED HYDROCARBON RESIDUES IN HUMAN ADIPOSE TISSUE  
 (mean of 168 Canadian Samples)<sup>1</sup>

<u>Compound</u>	<u>Mean Values in <math>\mu\text{g}/\text{kg}</math> Wet Weight</u>	<u>Percentage of Samples Containing Residues</u>
PCB	907	100
Hexachlorobenzene	62	100
$\gamma$ BHC (Lindane)	65	88
Oxychlorane	55	97
Trans-nonachlor	65	99
Heptachlor Epoxide	43	100
Dieldrin	69	100
p,p'-DDE	2095	100
o,p'-DDT	31	100
p,p'-TDE	6	26
p,p'-DDT	439	100

(Humphrey, undated). Of a group of 161 adult participants, 105 individuals were identified who consumed more Lake Michigan fish than recommended by the Michigan Department of Public Health (no more than one fish meal per week or 24 pounds per year). Medical histories, dietary records, and blood specimens were obtained for all participants. Humphrey reports that PCB blood levels ranged from 0.007 parts per million in persons who ate no fish, to a maximum of 0.366 parts per million for a person eating large quantities of Lake Michigan fish. The first of these data points corresponds rather closely to the upper levels of the unexposed persons, and the lower mean of exposure among Japanese involved in the Yusho incident. The second relates closely to the mean reported for individuals who were occupationally exposed (see Table 15).

The 1973 mean PCB blood value reported in the Humphrey study for persons consuming more than 24 pounds of sport fish from Lake Michigan was 0.073 parts per million, as compared with persons who annually ate six pounds or less, for whom circulating blood titers of PCB were 0.020 parts per million. The mean PCB blood level in persons who consumed no Great Lakes fish, and thus avoided exposure via this route, was 0.017 parts per million. The data for 1974 showed similar values for blood PCB levels among the various study groups. Humphrey concludes, "There was a direct relationship between the size and quantity of sport species of fish eaten and the PCB levels found in human blood."

Indicative of the persistence of these compounds as a function of the fact that they are but poorly metabolized, the findings of this study show that PCB blood levels in exposed individuals did not decline significantly when consumption was eliminated for up to nine months.

Dose, as contrasted with exposure, is calculated upon mass of contaminant per unit of body mass per unit of time. The Food and Drug Administration recommends that PCB dosage not exceed 1  $\mu\text{g}/\text{kg}$  body weight/day for long term exposure. In the Humphrey study, 82 percent of those in the high consumption groups (greater than 24 pounds of fish eaten per year) received a maximum annual dose exceeding Food and Drug Administration guidelines. Within this group, calculated dose ranges were from 0.49  $\mu\text{g}$  PCB/kg body weight/day to 3.94  $\mu\text{g}$  PCB/kg body weight/day, with a mean value of 1.70  $\mu\text{g}$  PCB/kg body weight/day.

#### Potential for Infant Exposure

In primate PCB exposure studies using rhesus monkeys, Allen and Barsotti (1976) have shown substantial exposure of infant progeny in utero as a result of transplacental passage of the PCB molecule. These findings substantiate the original suspicions of Hirayama (1976) who observed that babies born to Yusho mothers often had abnormal characteristics. Later, Kuwabara *et al.* (1978) reported blood levels for female workers exposed to PCBs and values for their newborn children. The blood range for 20 workers was 8.3 to 84.5 parts per billion, and that of 39 children born to these women was 0.8-93.2 parts per billion. A comparison of the mean values, 36.8 parts per billion and 14.3 parts per billion respectively, suggests that in the case of occupationally exposed women, children received an in utero exposure equivalent to nearly forty percent of their mother's total blood burden.

TABLE 15

PCBs IN HUMAN BLOOD OF PATIENTS WITH YUSHO,  
WORKERS ENGAGED IN THE PRODUCTION OF KANECHLOR 200-600,  
AND PERSONS NOT EXPOSED BY EITHER OF THESE ROUTES<sup>1</sup>

<u>Material Examined</u>	<u>PCB in <math>\mu\text{g}/\text{kg}</math> (ppb)</u>	
	<u>Mean</u>	<u>S.D.</u> <u>Range</u>
<u>WHOLE BLOOD</u>		
Yusho Patients	7.0	2.0 - 26.0
Non-Exposed	3.0	1.0 - 7.0
<u>PLASMA</u>		
Yusho Patients	6.3 $\pm$ 4.0	2.0 - 15.0
Non-Exposed	3.0 $\pm$ 1.3	1.0 - 7.0
<u>WHOLE BLOOD</u>		
Yusho Patients	4.8 $\pm$ 2.9	1.0 - 12.0
Non-Exposed	2.8 $\pm$ 1.5	1.0 - 6.0
<u>WHOLE BLOOD</u>		
Occupationally Exposed Workers <sup>2</sup>	364.0 $\pm$ 262.0	60.0 - 920.0

<sup>1</sup>Data after Kuratsune et al., 1976

<sup>2</sup>Ambient air concentrations 0.05-0.2 mg/m<sup>3</sup> PCBs Kanechlor 300 + 400 .

Abe et al. (1975) reported similar findings among children of mothers exposed to Yusho. These workers compared concentrations of PCBs in the plasma of 18 mothers with Yusho (range of 3.0 to 33.0 parts per billion observed) with their 30 offspring (range 1.0-20.0 parts per billion observed). When mean values from these data are compared  $11.2 \pm 7.32$  parts per billion and  $6.7 \pm 4.28$  parts per billion, respectively, it appears that infant values approach 60 percent of total maternal blood titers. It must be noted, however, that additional exposures from other exogenous sources may have some effect on these data.

Among non-occupationally exposed mothers, Kuwabara et al (1979) present data which suggests even higher values. For a child of less than one month of age, blood levels were found to be 96.5 percent of his mother's equivalent blood titer. For all children up to seven months of age, circulating blood titers were found to be 73.7 percent of their mother's total blood concentration. While the data unquestionably is affected by nursing, the values suggest substantial prepartum exposure of infants. Because of the lack of definitive data, however, it is fortuitous that additional studies are planned to fill existing knowledge gaps (Humphrey, 1980).

At parturition, infants may potentially be exposed to an additional source of PCBs, at levels substantially greater than those experienced in utero. Based on 1971-1972 data, Nisbet (1976) warned that, "...exposure of breast-fed infants is likely to be very much greater than that of any adult, even an adult who likes fish." Allen and Barsotti (1976) demonstrated the effects of exposure from nursing in rhesus monkeys. While absolute values of PCBs were apparently not measured in the maternal milk of rhesus monkeys, the rapid increase in PCB levels in the tissues of the infants was attributed to consumption of PCB contaminated milk from their mothers. Within 2 months following birth, the offspring exhibited the classic dermal symptoms of PCB intoxication, and within 8 months, 3 of the 6 infants died of PCB poisoning. These workers further note that the 3 surviving infants were removed from continuing dietary sources of PCB and subsequently showed marked improvement in their physical state.

At approximately the same time that the non-human primate data was made available, Grant et al. (1976) reported the values of Holdreient et al. (1975) on PCB residues in human milk from residents of Ontario, Canada. The values observed by these workers for the years 1969-1974 ranged between 0.7 and 3.0 mg/kg PCB on a fat basis with mean values of 1.0 mg/kg for 1969-70 and 1.2 mg/kg for 1971-72 and 1973-74.

In a subsequent study, Mes and Davies (1979) report an average of 12 ng/g PCB as Aroclor 1260 in whole milk samples from Canadian mothers. A maximum of 68 ng/g was observed in one sample, and a total of 98 percent of the mothers examined had residues equal to or greater than 1 ng/g on a whole milk basis. These workers also provide interesting time series data between the years 1967 and 1975 (see Table 16). For all organochlorine compounds other than PCB for which complete data sets exist, Mes and Davies show substantial declines through time. For PCB analyses, however, a doubling was observed between 1970 and 1975.

TABLE 16  
TREND OF ORGANOCHLORINE RESIDUES IN CANADIAN  
HUMAN MILK<sup>1</sup>

<u>Compound</u>	<u>Average ng/g Whole Milk</u> <u>Year of Sample Collection</u>		
	<u>1967</u>	<u>1970</u>	<u>1975</u>
PCB as Aroclor 1260	--	6	12
HCB	--	--	2
$\beta$ HCH	--	--	2
$\alpha$ HCH	3	2	--
Heptachlor Epoxide	3	4	1
Oxychlorthane	--	--	1
Trans-Nonachlor	--	--	1
p,p'-DDE	103	56	35
Dieldrin	5	5	2
o,p'-DDT	5	3	3
p,p'-TDE	4	3	--
p,p'-DDT	33	15	6

<sup>1</sup>From Mes and Davies (1979)

A comparison of the regional distribution of organochlorine residues in breast milk from Canadian mothers was also reported by these authors (Table 17). The geographic distribution of the residues observed in this study correlated well with observations of Grant *et al.* (1976) for distribution of PCBs in human adipose tissue. A comparison of Tables 13 and 17 demonstrate these relationships.

Studies of pooled breast milk samples by Westoo and Noren (1978) in Sweden show declines similar to the Mes and Davies data in other organochlorine compounds, but demonstrable increases in PCB residues between 1967 and 1977. The Swedish data reports 14 ng/g PCB on a fat basis for 1967, and a more than two fold increase by 1977 (30 ng/g PCB). The values for 1971 and 1974 were 26 and 24 ng/g, respectively. Of particular interest is the comment these authors make regarding the probable reason for this increase. They note that PCB levels appear to be increasing in spite of the restrictions imposed on the use of PCBs in 1972. They suggest that atmospheric fallout of PCB, "...make[s] a major contribution to the PCB levels in Swedish fish which are probably the main source of PCBs for the mothers."

Ames (1979) reporting the work of Harris and Highland (1977) listed values for organochlorine compounds in breast milk of 1400 women in the United States. The data from the Ames paper is shown in Table 18. While the mean of this study is significantly lower than that of either the Swedish or Canadian populations studied, the upper limit approaches the averages found by Mes and Davies (1979) and by Westoo and Noren (1978).

In a survey of children of occupationally exposed mothers, Kuwabara *et al.* (1979) reported an eight month old baby with a blood level of 115  $\mu\text{g}/\text{kg}$  PCB. An investigation revealed that this level apparently resulted from an exposure to contaminated breast milk of six months duration. This lead these authors to examine the effects of nursing upon children of non-occupationally exposed mothers. It was found that in 9 of the 17 children examined, their circulating titer of blood PCB was higher than that of their mothers. The mean value reported for the mothers was  $2.8 \pm 0.8$  ng/g PCB and the mean value for the blood of the children was  $3.8 \pm 3.6$  ng/g. Kuwabara and his associates also calculated daily exposure rates for infants based on these exposure levels. These authors assumed that a baby ingests approximately 1 kg of breast milk per day contaminated with an average of 30 parts per billion of PCB, they concluded that the average breast fed Japanese baby would receive a daily exposure of 30  $\mu\text{g}$  of PCB. Calabrese and Sorenson (1977) note that such exposure at the embryonic, fetal and neonatal ages is particularly hazardous, since often children of this age lack the liver microsomal enzyme systems to detoxify various natural and foreign chemicals, including those of a biphenolic nature.

Among individuals consuming Lake Michigan fish who participated in the Michigan blood level PCB study, Humphrey (undated) reports a single value for breast milk. He notes that the sample was received from a mother in the low exposure class, who had consumed 28 pounds of Lake

TABLE 17

 REGIONAL DISTRIBUTION OF ORGANOCHLORINE RESIDUES  
 IN CANADIAN HUMAN MILK<sup>1</sup>

<u>Compound</u>	<u>Regional Averages in ng/g whole milk</u>				
	<u>Eastern</u>	<u>Quebec</u>	<u>Ontario</u>	<u>Central</u>	<u>Western</u>
PCB, as Aroclor 1260	8	10	17	8	15
HCB	1	1	2	1	4
βHCH	1	1	3	2	2
Heptachlor Epoxide	1	1	1	1	2
Oxychlorane	1	1	1	1	1
Trans-Nonachlor	1	1	1	1	1
p,p'-DDE	29	34	34	21	59
Dieldrin	2	1	2	1	2
o,p'-DDT	3	1	6	-	1
p,p'-DDT	5	7	6	5	8

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<sup>1</sup>From Mes and Davies (1979)

TABLE 18  
 ORGANOCHLORINE CONTAMINANTS IN THE MILK OF 1400 WOMEN<sup>1</sup>

Compound	Number positive (%)	Mean of positives ( $\mu\text{g}/\text{kg}$ fat)*	Maximum ( $\mu\text{g}/\text{kg}$ fat)
DDE	100	3521	214,167
DDT	99	529	34,369
Dieldrin	81	164	12,300
Heptachlor epoxide	64	91	2,050
Oxychlorane	63	96	5,700
$\beta$ BHC	87	183	9,217
PCBs	30	2076	12,600

\*4.5 percent = mean fat content. †99 percent detectable  
 PCBs; (30 percent =  $>1100 \mu\text{g}$  per kilogram of fat, 1038 women,  
 Environmental Protection Agency, 1977).

<sup>1</sup>From Ames (1979)

Michigan fish during a two year period, 1973-74, approximately one-half of the recommended maximum of 28 pounds per annum. This woman had a circulating blood titer of 0.053 parts per million PCB. A breast milk sample collected the same month as the blood sample contained 4.0 mg/kg PCB on a fat basis (2% fat content), equivalent to 80.0 ng/g PCB on a whole milk basis. This value is substantially above the levels reported in the literature for the general population and for the portion of Michigan residents unexposed to large quantities of Lake Michigan fish (Humphrey, 1980). It was further suggested that on the basis of present work, the value reported for this sample is not unusual for breast milk from women exposed to substantial quantities of Lake Michigan fish.

Applying the reasoning of Kuwabara *et al.* (1979) to the values from this sample, it can be reasonably assumed that a nursing infant will consume 1 kg of breast milk per day. At a level of 80 ng/g, the child is exposed to 80  $\mu$ g of PCB each day.

As was previously noted, dose, as contrasted with exposure, is calculated upon mass of compound per mass of body weight per unit of time. To carry the analogy of exposure to dose, it is necessary to make only one further assumption, i.e., that a reasonable birth weight is 3-4 kg (approximately 6-8 pounds). Thus, the dose rate for a child of these proportions would be between 20 and 26.6  $\mu$ g/kg body weight/day, some 20 to 25 times the maximum daily dose rate of 1  $\mu$ g/kg/day PCB recommended by the U.S. Food and Drug Administration for adult intake.

#### Other Health Related Studies

Internal pool mechanisms are undoubtedly active, but their long term importance is, at present, a matter of conjecture. Calabrese and Sorenson (1979) have reported that five percent of mothers of normal infants secrete milk which inhibits the activity of glucuronyl transferase by means of the action of a steroid present in their breast milk. This tends to suppress or inhibit elimination of bilirubin, and can be expected to reduce the ability of about 5 percent of breast fed children to eliminate PCBs.

Another internal mechanism for which no data apparently exist relates to the question of mobilization of lipid resources during starvation or normal growth cycles. As a child undergoes natural growth and developmental cycles, he is repeatedly subjected to intensive spurts of growth in which much of the available fatty materials are drawn into circulation and metabolized to provide energy for the increased state of anabolic activity. It is to be expected that stored organochlorine contaminants in body fat may also be mobilized during these periods providing an internal re-exposure mechanism. For the most part, these intensive bursts of growth occur during periods when unprecedented growth and maturation of tissues, long bone development, and myelination of the central nervous system is occurring. The long term effect of these processes, if any, is unknown.

Data on tissue and organ level effects and upon cellular impacts is also available. While a complete discussion is beyond the extent of

this paper, it is worth acknowledging representative efforts in this area. Of potential great interest to high risk segments of the population, including persons consuming excessive quantities of Great Lakes fish, are the findings of Dougherty *et al.* (1980) relative to alteration of human sperm densities as a result of exposure to toxic substances. Allen *et al.* (1976) report liver hypertrophy and hepatocellular degeneration in rats exposed to PCBs. Loose *et al.* (1977) report that PCB and other organochlorine compounds induce humoral immunosuppression, and Stotz and Greichus (1978) note abnormalities in the ultrastructure of liver hepatocytes. Using combinations of pesticides and PCBs, Parkki *et al.* (1977) report alterations of enzyme activities in rats. Ohnishi and Noda (1977) have demonstrated a range of cellular response to PCB in conjunctival cells from surface roughening of the endoplasmic reticulum to death of the cells.

Of particular interest may be the relationship of PCB and other organochlorine compounds to the alteration of disease states. Nobuyuki *et al.* (1973) have demonstrated that PCBs promote the induction of chemically caused neoplasms by benzene hexachloride in mice, and Loose *et al.* (1978) have found that PCB impairs murine resistance to both malaria (Plasmodium berghei) and the gram negative endotoxin of Salmonella typhosa.

#### Summary

A conceptual framework for an ecosystem approach to the question of the impact of residue forming toxic substances of anthropogenic origin in the Great Lakes has been presented. This framework is an attempt to link a definite and orderly progression of events related to input sources of toxic substances and their compartmentalization phenomena. It is believed to be of utility because it considers interrelationships and properties between separate entities, incorporates defined criteria, and utilizes a series of linkages into which quantifiable values and numerically precise data can be inserted to enable an adequate mechanism for understanding the fate, transport and effects of toxic organochlorine compounds.

Given the magnitude of the Great Lakes ecosystem in both spatial and temporal scales, the interdependency of the various ecosystem compartments and the multiplicity of exogenous substances with differing chemical/biological reactions, it is difficult to conceive that any other approach to the question could adequately address the complexity of the issue. While short term laboratory tests are useful in delineating areas of the environment where problems can be anticipated, often the question of scale makes direct translation of results to the Great Lakes difficult, if not impossible. The approach described in this effort is an attempt to overcome these difficulties for toxic xenobiotic organochlorine compounds.

## REFERENCES

- Abe, S., Inoue, Y., and Takamatsu, M. 1975. Polychlorinated biphenyls in plasma of Yusho children born to mothers who had consumed oil contaminated by PCB. Fukuoka Acta Med. 66: 605-609 (in Japanese).
- Allen, J.R. and Barsotti, D.A. 1976. The effects of transplacental and mammary movement of PCBs on infant rhesus monkeys. Toxicology 6: 331-340.
- Allen, J.R., Carstens, L.A., and Abrahamson, L.J. 1976. Responses of rats exposed to polychlorinated biphenyls for fifty-two weeks. I. Comparison of tissue levels of PCB and biological changes. Arch. Environ. Contam. Toxicol. 4(4): 404-419.
- Allen, J.R., Carstens, L.A., and Barsotti, D.A. 1974. Residual effects of short term low level exposure of nonhuman primates to polychlorinated biphenyls. Toxicol. Appl. Pharmacol. 30: 440-451.
- Ames, B. 1979. Identifying environmental chemicals causing mutations and cancer. Science 204: 587-593.
- Andren, A. and Doskey, P. 1978. Data on PCB concentrations in the atmosphere and dry deposition. Presented at a Workshop on the transport of fate of Polychlorinated Biphenyls in the Great Lakes, April 3-4, 1978, Grosse Ile, Michigan.
- Armstrong, D. 1980. Personal communication.
- Benvenue, A., Ogata, J.N., and Hylin, J.W. 1972. Organo-chlorine pesticides in rainwater, Oahu, Hawaii, 1971-72. Bull. Env. Cont. Toxic. 8: 238-241.
- Bidleman, T.F. and Christensen, E.J. 1979a. Aerial deposition of chlorinated hydrocarbons in urban and coastal South Carolina. Presented at September 1979 meeting of the American Chemical Society, Washington, D.C.
- Bidleman, T.F. and Christensen, E.J. 1979b. Atmospheric removal processes for high molecular weight organochlorines. J. Geophysical Res. 84: 7857-7862.
- Bidleman, T.F. and Olney, C.E. 1974. Chlorinated hydrocarbons in the Sargaso Sea atmosphere and surface water. Science 183: 516-518.
- Bierman, V.J. and Swain, W.R. 1978. DDT loss rates in low lipid fishes of the Great Lakes. International Association for Great Lakes Research, Abstracts of Papers, Twenty First Conference on Great Lakes Research. p. 12.

- Biros, F.J., Walker, A.C. and Medberry, A. 1970. Polychlorinated Biphenyls in human adipose tissue. Bull. Environ. Contamin. and Toxicol. 5: 317-323.
- Bowes, G.W. and Jonkel, C.J. 1975. Presence and distribution of Polychlorinated Biphenyls (PCB) in arctic and subarctic food chains. J. Fish Board Can. 32: 2111-2123.
- Calabrese, E.J., and Sorenson, A.J. 1977. The health effects of PCBs with particular emphasis on human high risk groups. Reviews on Environmental Health 2(4): 285-304.
- Clausen, J., Braestrup, L. and Berg, O. 1974. The content of polychlorinated hydrocarbons in arctic mammals. Bull. Env. Cont. and Toxic. 12: 529-534.
- Dennis, D.S. 1976. Polychlorinated Biphenyls in the surface waters and bottom sediments of the major drainage basins of the United States. Proceedings of the National Conference on Polychlorinated Biphenyls, Nov. 19-21, 1975, Chicago, Illinois. NTIS PB-253-248: 183-194.
- DiToro, D.M. 1974. Vertical interactions in phytoplankton models - an Eigenvalue Analysis. International Association for Great Lakes Research 17th Conference on Great Lakes Research, Abstracts: 16.
- Doskey, P.V. 1978. Transport of Airborne PCBs to Lake Michigan. Master of Science Thesis, University of Wisconsin - Madison. pp. 185.
- Dougherty, R.C., Whitaker, M.J., Tang, S., Boitcher, R., Keller, M., and Kuehl, D. 1979. Sperm density and toxic substances: a potential key to environmental health hazards. In press.
- Eisenreich, S.J. 1980. Personal communication.
- Eisenreich, S.J. and Johnson, T. 1980. Personal communication
- Eisenreich, S.J., Hollod, G.J. and Johnson, T.C. 1979. Accumulation of PCBs in surficial Lake Superior sediments: atmospheric deposition. Environ. Science and Technol. 13: 569-573.
- Eisenreich, S.J., Hallod, G.J., Johnson, T.C. and Evans, J. 1979. Polychlorinated Biphenyl and other microcontaminants--sediment interactions in Lake Superior. Processes Involving Contaminants and Sediments, ed. R.A. Boher, Ann Arbor Press.
- Eisenriech, S.J., Looney, B.B. and Thornton, J.D. 1980 Assessment of Airborne organic contaminants in the Great Lakes ecosystem. A report to the Ecological and Geochemical Expert Committee and the Science Advisory Board of the International Joint Commission (typescript) pp. 48 and appendices.

- Erstfeld, K.M., Rice, C.P. and Meyers, P.A. 1980. The surface micro-layer: A postulated agent in the air/water exchange of PCBs and other related pollutants. International Association for Great Lakes Research, 23rd Conference on Great Lakes Research, Abstracts: 47.
- Ewing, B.B. 1977. Monitoring to detect previously unrecognized pollutants in surface waters. USEPA Report No. 560/6-77-015.
- Finklea, J., Priester, L.E., Creason, J.P., Hauser, T., Hinners, T. and Hammer, D.I. 1972. Polychlorinated Biphenyl residues in human plasma expose a major urban pollution problem. Amer. J. Pub. Health 62(5): 645-651.
- Fox, M.E. 1978. Pentachlorophenol in the Great Lakes Basin. Presented at the 21st Conference on Great Lakes Research, International Association for Great Lakes Research, Windsor, Ontario.
- Frank, R., Braun, H.E., Sirons, G., Van Hove Holdrinet, M., and Ripley, B.D. 1978. Stream flow quality-pesticides in eleven agricultural watersheds in Southern Ontario, Canada 1974-77. Task Group C (Canadian section) Activity 1. International Reference Group on Great Lakes Pollution from land use activities. International Joint Commission. pp. 174.
- Frank, R., Thomas, R.L., Holdrinet, M., Kemp, A.L.W. and Braun, H.E. 1979. Organochlorine insecticides and PCB in surficial sediments (1968) and sediment cores (1976) from Lake Ontario. J. Great Lakes Res. 5(1): 18-27.
- Giam, C.S., Wong, M.K., Hanks, A.R., and Sackett, W.M. 1973. Chlorinated hydrocarbons in plankton from the Gulf of Mexico and Northern Caribbean. Bull. Env. Cont. and Toxic. 11: 376-382.
- Gillett, J.W., Hill, J., Jarvinen, A.W., Schoor, W.P. 1974. A conceptual model or the movement of pesticides through the environment: a contribution of the EPA alternative chemicals program. USEPA Ecological Res. Series. EPA-660/3-74-024. pp. 79.
- Glooschenko, W., Strachan, W.M.J., and Sampson, R.C.J. 1976. Distribution of pesticides and polychlorinated biphenyls in water sediments and seston of the Upper Great Lakes - 1974. Pestic. Monit. J. 10(2): 61-67.
- Grant, D., Mes, J. and Frank, R. 1976. PCB residues in human adipose tissue and milk. National Conference on Polychlorinated Biphenyls, November 19-21, 1975, Chicago, Illinois. NTIS PB-253-248. pp. 144-146
- Great Lakes Research Institute. 1973. Selected analysis and monitoring of Lake Erie water quality - Annual Report submitted to Erie County Health Department of the Commissioners of Erie County, P.R. in International Joint Commission (1978).

- Haile, C.M., Veith, G.D., Lee, G.F. and Boyle, W.C. 1975. Chlorinated Hydrocarbons in the Lake Ontario ecosystem (IFYGL). USEPA Ecological Research Series. EPA-660/3-75-022. pp. 28.
- Harris, S.G. and Highland, J.H. 1977. Birthright Denied. Environmental Defense Fund, Washington, D.C.
- Herdendorf, C.E., Cooper, C.L. and Gessner, M. 1978. Lake Erie water quality status report for 1977-nearshore. Prepared for USEPA Region V. Ohio State University Center for Lake Erie Area Research. Technical Report No. 80.
- Higuchi, K. ed. 1976. PCB poisoning and pollution. Kodansha Ltd/Academic Press/New York. pp. 184.
- Hirayama, C. 1976. Clinical aspects of PCB poisoning in PCB Poisoning and Pollution. Higuchi, ed. Kodansha Ltd/Academic Press. New York. pp. 87-104.
- Holdrient, M. Braun, H.E., Frank R., Stopps, G.J. and McWade, J.W. 1975. Unpublished results.
- Hollod, G.J. 1979. Polychlorinated Biphenyls (PCBs) in the Lake Superior ecosystem: Atmospheric deposition and accumulation in the bottom sediments. Ph.D. Thesis, University of Minnesota. pp. 247.
- Humphrey, H.E.B. Undated. Evaluation of changes of the level of Polychlorinated Biphenyls (PCB) in human tissue. Final Report on FDA Contract 223-73-2209. Michigan Department of Public Health, Lansing, Michigan. pp. 591 Appendices.
- Humphrey, H.E.B. 1980. Personal communication.
- Ingersoll, B. 1977. The miracle that go away. Chicago pp. 100-125 (not inclusive).
- International Joint Commission 1975. Water quality objectives sub-committee report: Persistent compounds. Water Quality Board Report (1974) to the International Joint Commission. Appendix A. pp. 123 and Appendices.
- International Joint Commission. 1977. Great Lakes Water Quality. Status Report on the persistent toxic pollutants in the Lake Ontario Basin by the Implementation Committee. pp. 95.
- International Joint Commission. 1978. Great Lakes Water Quality. Status report on organic and heavy metal contaminants in the Lakes Erie, Michigan, Huron and Superior Basins. Great Lakes Water Quality Board Appendix E. pp. 373.

- Jelinik, C.F., and Corneliussen, P.E. 1976. Levels of PCBs in the U.S. food supply. National Conference on Polychlorinated Biphenyls, November 19-21, 1975, Chicago, Illinois. NTIS PB-253-248. pp. 147-154.
- Kang, S.W. and Lick, W. 1980. The entrainment and deposition of fine-grained lake sediments. International Association for Great Lakes Research, 23rd Conference on Great Lakes Research, Abstracts: 29.
- Kemp, A.L.W. and Harper, N.S. 1977. Sedimentation rates in Lake Huron and Georgian Bay. J. Great Lakes Res. 3(3-4): 215-220.
- Kemp, A.L.W., MacInnis, G.A., and Harper, N.S. 1978. Sedimentation rates and sediment budget for Lake Erie. J. Great Lakes Res. 3: 221-233.
- Kimbrough, R.D. 1976. Pathological findings associated with chronic experimental exposure to PCBs. National Conference on Polychlorinated Biphenyls, November 19-21, 1975, Chicago, Illinois. NTIS PB-253-248. pp. 30-34.
- Kimbrough, R.D., Linder, R.E., Burse, V.W. and Jennings, R.W. 1973. Adenofibrosis in the rat liver. Arch. Environ. Health 27: 390-395.
- Kinkead, J.D. and Chatterjee, R.M. 1974. A limnological survey of nearshore waters of Lake Superior. Proc. 17th Conf. Great Lakes Res., Internat. Assoc. Great Lakes Res. pp. 549-573.
- Kinkead, J.D. and Hamdy, Y. 1978. Trends in the mercury content of Western Lake Erie fish and sediment 1970-77. Ontario Ministry of the Environment. pp. 19.
- Kleinert, S.J. 1976. The PCB problem in Wisconsin. A report prepared for the Joint Hearing of the Assembly. Environmental Quality Committee with the Senate and Assembly Natural Resources Committies on NR 212 administrative rules for PCB effluent standards.
- Kleinert, S.J. 1976. Sources of Polychlorinated Biphenyls in Wisconsin. National Conference on Polychlorinated Biphenyls, November 19-21, 1975, Chicago, Illinois. U.S. Department of Commerce. NTIS PB-253 248. pp. 124-126.
- Kolbye, A.C. 1972. Food exposures to Polychlorinated Biphenyls. Environmental Health Persp. 1: 85-88.
- Kuwabara, K., Yakushiji, T., Watanabe, I., Yoshida, S., Koyama, K., Kunita, N. and Hara, I. 1978. Relationship between breast feeding and PCB residues in blood of the children whose mothers were occupationally exposed to PCBs. Int. Arch. Occup. Environ. Hlth. 41: 189-197.

- Kubawbara, K., Yakushiji, T., Wantanabe, I., Yoshida, S., Koyama, K., and Kunita, N. 1979. Levels of Polychlorinated Biphenyls in blood of breast-fed children whose mothers are non-occupationally exposed to PCBs. Bull. Environ. Contam. Toxicol. 21: 458-462.
- Kuwabara, K., Yakushiji, T., Watanabo, I., Yoshida, S., Yoyama, K., and Kunita, N. 1979. Increase in the human blood PCB levels promptly following ingestion of fish containing PCBs. Bull. Environ. Contam. Toxicol. 21: 273-278.
- Kuratsune, M., Masuda, Y. and Nagayama, J. 1976. Some recent findings concerning Yusho. National Conference on Polychlorinated Biphenyls, November 19-21, 1975, Chicago, Illinois. NTIS PB-253-248. pp. 14-29.
- Kuratsune, M., Yoshimura, T., Matsuzaka, J. and Yamaguchi, A. 1972. Epidemiologic study on Yusho, a poisoning caused by ingestion of rice oil contaminated with a commercial brand of Polychlorinated Biphenyls. Envir. Hlth. Perspect. 1: 119.
- Kutz, F.W. and Strassman 1976. Residues of Polychlorinated Biphenyls in the general population of the United States. National Conference on Polychlorinated Biphenyls, November 19-21, 1975, Chicago, Illinois. NTIS PB-253-248. pp. 139-143.
- Lake Michigan Enforcement Conference. 1972. An evaluation of DDT and Dieldrin in Lake Michigan. USEPA Ecological Res. Series. EPA-R3-72-003. pp. 139.
- Lick, W. 1979. Transport of contaminants in the Great Lakes. Department of Mechanical and Environmental Engineering, University of California, Santa Barbara, California. Typescript pp. 53.
- Loose, L.D., Pittman, K.A., Benitz, K.F. and Silkworth, J.B. 1977. Polychlorinated Biphenyl and Hexachlorobenzene induced humoral immuno suppression. J. Reticuloendothelial Soc. 22(3): 253-271.
- Loose, L.D., Silkworth, J.B., Pittman, K.A., Benitz, K.F., and Mueller, W. 1978. Impaired host resistance to endotoxin and malaria in Polychlorinated Biphenyl - and Hexachlorobenzene - treated mice. Infection and Immunity 20(1): 30-35.
- McGrath, M. 1980. The fourth coast. EPA Journal 6(5): 22-24.
- McNaught, D.G., Griesmer, D.A., and Kennedy, M. 1978. Acute effects of PCBs on fluxes of carbon in the Lake Huron ecosystem. International Association for Great Lakes Research, Abstracts: 99.
- McNaught, D.C. 1980. Personal communication.
- Mes, J. and Davies, D. 1979. Presence of Polychlorinated Biphenyl and organochlorine pesticide residues and the absence of Polychlorinated Terpehnyls in Canadian human milk samples. Bull. Environ. Contamn. Toxicol. 21: 381-387.

- Mes, J., Campbell, D.S., Robinson, R.N. and Davies, I.J.A. 1977. Polychlorinated Biphenyl and organochlorine pesticide residues in adipose tissue of Canadians. Bull. Env. Contam. and Toxicol. 17: 196-203.
- Metcalf, R.L., Sanborn, J.R., Lu, P. and Nye, D. 1975. Laboratory Model ecosystem studies of the degradation and fate of radiolabeled tri-tetra-and pentachlorobiphenyl compared with DDE. Arch. Env. Cont. and Toxic. 3(2): 151-165.
- Michigan Department of Natural Resources 1978. STORET retrieval-Lake Superior in International Joint Commission (1978).
- Mount, D.I. 1976. Summary of water analysis persistent to the barrels in Lake Superior Environmental Research Laboratory-Duluth; Press release.
- Mullin, M.D. and Filkins, J.C. 1980. Variations in PCB distributions in precipitation, Saginaw Bay water and Saginaw Bay fish utilizing high resolution glass capillary gas chromatography. International Association for Great Lakes Research, 23rd Conference on Great Lakes Research, Abstracts: 46.
- Murphy, T.J. 1978. Polychlorinated Biphenyls in precipitation in the Lake Michigan basin. USEPA Ecological Research Series. EPA-600/3-78-071. pp. 39.
- Murphy, T.J. 1980. Personal communication.
- Murphy, T.J. and C.P. Rzeszutko. 1977. Precipitation Inputs of PCBs to Lake Michigan. J. Great Lakes Res. 3(3-4): 305-312.
- Nagayama, J., Masuda, Y. and Kuratsune, M. 1975. Chlorinated dibenzofurans in kanechlors and rice oils used by patients with Yusho. Fukuoka Acta. Med. 66(10): 593-599.
- Nagayama, J., Masuda, Y., and Kuratsune, M. 1977. Determination of Polychlorinated Dibenzofurans in tissues of patients with Yusho. Fd. Cosmet. Toxicol. 15: 195-198
- Nisbet, I.C.T. 1976. Environmental Transport and occurrence of PCBs in 1975. National Conference on Polychlorinated Biphenyls, November 19-21, 1975, Chicago, Illinois. NTIS PB-253-248. pp. 254-256.
- Nobuyuki, I., Nagasaki, H. and Arai, M. 1973. Histopathologic studies on liver tumorigenesis induced in mice by technical Polychlorinated Biphenyls and its promoting effect on liver tumors induced by benzene hexachloride. J. Natnl. Cancer Inst. 51: 1637.
- Nriagu, J.O., Kemp, A.L., Wong, H. and Harper, N. 1979. Sedimentary record of heavy metal pollution in Lake Erie. Geochem. Cosmochim. Acta 43: 247-258.

- Ohnishi, Y. and Noda, S. 1977. Effects of Polychlorinated Biphenyls (PCB) on dissociated conjunctival cells in vitro. Japanese Journal of Ophthalmology 21: 496-506.
- Ontario Ministry of the Environment 1976. Polychlorinated Biphenyls in the Ontario Environment, p. 23 in Great Lakes Water Quality - Appendix E - Status Report on the persistent toxic pollutants in the Lake Ontario Basin. International Joint Commission.
- Parkki, M.G., Marniemi, J., Vainio, H. 1977. Longterm effects of single and combined doses of DDT and PCB on drug-metabolizing enzymes in rat liver. J. Toxicol. Environ. Health 3: 903-911.
- Peel, D.A. 1975. Organochlorine residues in antarctic snow. Nature 154: 324-325.
- Pennsylvania Department of Natural Resources 1977. PCBs in Pennsylvania Waters. Bureau of Water Quality Management. Publication No. 51. pp. 86.
- Peterle, T.J. 1969. DDT in Antarctic snow. Nature 224: 620.
- Price, H.A. and Welch, R.L. 1972. Occurance of Polychlorinated Biphenyls in humans. Environ. Health Perspectives 1: 73-78.
- Rainey, R.H. 1967. Natural displacement of pollution from the Great Lakes. Science 155: 1242-1243.
- Rhee, G. 1980. Personal communication.
- Rice, C. 1980. Personal communication.
- Richardson, W.L., Smith, E. and Filkins, J. 1980. Distribution of Aroclor 1254 in Saginaw Bay during 1977. International Association for Great Lakes Research, 23rd Conference on Great Lakes Research, Abstracts: 47.
- Robbins, J.A., Edgington, D.N., and Kemp, A.L. 1978. Comparative Pb-210, CS-137, and pollen geochronologies of sediments from Lake Ontario and Lake Erie. Quat. Res. 10: 256-278.
- Robbins, J.A. and Edgington, D.N. 1975. Determination of recent sedimentation rates in Lake Michigan using Pb-210 and Cs-137. Geodium. Cosmocluism Acta. 39: 285-304.
- Robbins, J.A. 1978. Geochemistry and geophysical applications of radioactive lead. Biochemistry of Lead in the Environment Vol. 1. ed. J.O. Nriagu. pp. 258-408.

- Sanderson, M. 1977. Agricultural watershed studies in the Canadian Great Lakes Drainage Basin - Precipitation, Quantity and Quality. Task Group C (Canadian section) Activity 1. International Reference Group on Great Lakes Pollution from Land Use Activities, International Joint Commission. pp. 137.
- Schacht, R.A. 1974. Pesticides in the Illinois waters of Lake Michigan. USEPA Ecological Res. Series. EPA-660/3-74-002. pp. 55.
- Sly, P.G. and Sandilands, R.G. 1980. Local variations in Lake sediment composition and characteristics. International Association for Great Lakes Research, 23rd Conference on Great Lakes Research, Abstracts: 28.
- Smith, S.E., Sonzogni, W.C., Simmons, M.S. and Rice, C.P. 1980. Organic and heavy metal contaminants data review - a case study - Lake Michigan. International Association for Great Lakes Research, Abstracts: 49.
- Sonzogni, W.C., Monteith, T.J. and Skimin, W.E. 1980. Critical assessment of U.S. land derived pollutant loadings for the Great Lakes. International Association for Great Lakes Research, 23rd Conference on Great Lakes Research, Abstracts: 37.
- Stalling, D. 1980. Personal communication.
- Stanley, C.W., Barney, J.E., Hetton, M.R. and Yobs, A.R. 1971. Measurement of atmospheric levels of pesticides. Environ. Sci. Technol. 5(5): 430-435.
- Stotz, I.J., Greichus, Y.A. 1978. The effects of a Polychlorinated Biphenyl (Aroclor 1254) on the white pelican: Ultrastructure of hepatocytes. Bull. Env. Contamin. Toxicol. 319-325.
- Strachan, W.M.J. 1976. Chloroform extractable organic compounds in the International Great Lakes. Identification and Analysis of Organic Pollutants in Water, L.H. Keith, ed. Ann Arbor Science Press, Inc. pp. 479-497.
- Strachan, W.M.J. 1978. Unpublished results reported in International Joint Commission 1978.
- Strachan, W.M.J. and Glass, G.E. 1978. Organochlorine substances in Lake Superior. J. Great Lakes Res. 4(3-4): 389-397.
- Strachan, W.M.J. and Haneault, H. 1979. Polychlorinated Biphenyls and organochlorine pesticides in Great Lakes precipitation. J. Great Lakes Res. 5(1): 61-68.
- Swain, W.R. 1978. Chlorinated organic residues in fish, water and precipitation from the vicinity of Isle Royale, Lake Superior. J. Great Lakes Res. 4(3-4): 398-407.

- Swain, W.R. 1980. Unpublished data.
- Swain, W.R., Wilson, R.M., Neri, V.P. and Porter, G.S. 1975. Evaluation of the effects of a harbor bubbler system for winter navigation on the water quality of Howard's Bay in the Duluth-Superior Harbor-Winter 1974-75. Final report to the United States Army Corps of Engineers, St. Paul District Office. pp. 66 and Appendices.
- Sydor, M., Stortz, K.R. and Swain, W.R. 1978. Identification of contaminants in Lake Superior through LANDSAT I DATA. J. Great Lakes Res. 4(2): 142-148.
- Torrey, M.S. 1976. Environmental Status of the Lake Michigan Region- Volume 3. Chemistry of Lake Michigan. Argonne National Laboratory, Argonne, Illinois. pp. 418.
- Thomann, R.V. 1978. Size dependent model of hazardous substances in aquatic food chain. USEPA Ecological Research Series. EPA-600/3-78-036. pp. 39.
- Thomann, R.V. 1980. Steady state model of fate of chemicals in diverse aquatic food chains. Manhattan College, Bronx, New York (Typescript) pp. 46.
- Ulanoff, D.M., Hunt, G.E., Pirbazari, M. and Webber, W.J. 1980. Methodologies and analytical procedures for investigation of sorption reactions of Polychlorinated Biphenyls in aquatic environments. International Association for Great Lakes Research, 23rd Conference Great Lakes Research, Abstracts: 46.
- United States Fish and Wildlife Service. National pesticide monitoring program - Fish 1974. Fall 1974 analytical results.
- Upper Lakes Reference Group, International Joint Commission. 1977. The waters of Lake Huron and Lake Superior, Volume III - Parts A and B. Lake Superior.
- Veith, G.D. 1973. Chlorinated hydrocarbons in fish from Lake Michigan USEPA Water Quality Office, Project No. 16020PBE. pp. 129.
- Veith, G.D., Kuehl, D.W., Puglisi, F.A., Glass, G.E. and Eaton, J.G. 1977. Residues of PCBs and DDT in the Western Lake Superior ecosystem. Arch. of Env. Cont. and Toxic. 5: 487-499.
- Weininger, D. 1978. Accumulation of PCBs by Lake Trout in Lake Michigan. Ph.D. Thesis, University of Wisconsin-Madison. pp. 232.
- Westoo, G. and Noren, K. 1978. Human milk, Stockholm 1967-1977. Ambio 7 (2): 62-64.

Willford, W.A., Hesselburg, R.J. and Nicholson, L.W. 1976. Trends of Polychlorinated Biphenyls in three Lake Michigan Fishes. National Conferences on Polychlorinated Biphenyls, November 19-21, 1975, Chicago, Illinois. NTIS PB-253-248: 177-181.

Yobs, A.R. 1972. Levels of Polychlorinated Biphenyls in adipose tissue of the general population of the nation. Environ. Health Perspectives 1: 79-81.

