Related Pergamon Titles of Interest

Books

ALBAIGES:
Analytical Techniques in Environmental Chemistry (I and II)

HUTZINGER:
Aquatic Pollutants (Transformation and Biological Effects)
Chlorinated Dioxins and Related Compounds (Impact on the Environment)

STUCKEY & HAMZA:
Management of Industrial Wastewater in Developing Nations

VOWLES & CONNELL:
Experiments in Environmental Chemistry

WHO/UNEP:
Waste Discharge into the Marine Environment (Principles and Guidelines for a Mediterranean Action Plan)

ZOETEMAN:
Sensory Assessment of Water Quality

Journals

Water Research
Water Science and Technology
Water Supply and Management

Full details of all Pergamon publications/free specimen copy of any Pergamon journal available on request from your nearest Pergamon office.

BIOLOGICAL MONITORING IN WATER POLLUTION

JOHN CAIRNS, Jr.

and collaborators

A. L. Buikema, Jr.
D. S. Cherry
E. E. Herricks
R. A. Matthews
B. R. Niederlehner
J. H. Rodgers, Jr.
W. H. van der Schalie

PERGAMON PRESS

OXFORD · NEW YORK · TORONTO · SYDNEY · PARIS · FRANKFURT

U.K.
Pergamon Press Ltd., Headington Hill Hall,
Oxford OX3 0BW, England

U.S.A.
Pergamon Press Inc., Maxwell House, Fairview Park,
Elmsford, New York 10523, U.S.A.

CANADA
Pergamon Press Canada Ltd., Suite 104,
150 Consumers Rd., Willowdale, Ontario M2J 1P9, Canada

AUSTRALIA
Pergamon Press (Aust.) Pty. Ltd., P.O. Box 544,
Potts Point, N.S.W. 2011, Australia

FRANCE
Pergamon Press SARL, 24 rue des Ecoles,
75240 Paris, Cedex 05, France

FEDERAL REPUBLIC
OF GERMANY
Pergamon Press GmbH, 6242 Kronberg-Taunus,
Hammerweg 6, Federal Republic of Germany

Copyright © 1982 International Association on Water
Pollution Research & Control
and Pergamon Press Ltd.

All Rights Reserved. No part of this publication may be reproduced, stored in a retrieval system or transmitted in any form or by any means: electronic, electrostatic, magnetic tape, mechanical, photocopying, recording or otherwise, without permission in writing from the copyright holders.

First edition 1982

ISBN 0 08 028730 1

Originally printed in *WATER RESEARCH* (Vol. 14, No. 9, Vol. 16, Nos. 2, 3, Vol. 15, No. 8). Part I includes a new supplementary section.

Printed in Great Britain by A. Wheaton & Co. Ltd., Exeter

This book is dedicated to

Dr RUTH PATRICK

Senior Curator, Academy of Natural Sciences of Philadelphia
who was practising biological monitoring
before the term became fashionable

FOREWORD

This book on Biological Monitoring was planned so that individual self-contained parts on various aspects of the subject of Biological Monitoring could appear as review articles in the international technical journal *Water Research* (published by Pergamon Press for IAWPRC). The advantages of this arrangement were that subscribers to *Water Research* and members of the International Association on Water Pollution Research and Control, IAWPRC (formerly the International Association on Water Pollution Research, IAWPR), received parts of the book as soon as they were available, but it also allowed the costs of the book to be kept to the minimum by using the same text for both the review articles and the book.

This latter advantage causes a slight disadvantage which it is hoped will not inconvenience readers: since the parts in *Water Research* did not appear in sequence and were spread over several of its issues, the pages in the book are not numbered in proper sequence. However, the Table of Contents in the book gives the first and last pages for each part and in the text the parts are separated by a contrasting inserted page giving the part number and title. Each item is listed in the index by part number and page number within that part.

The information given in Part I has been expanded and updated by a supplementary section written nearly two years after the original part first appeared. This supplement is printed immediately after the original part. Please note that although a Part IIA appeared there will be no Part IIB.

S. H. JENKINS
Executive Editor, IAWPRC

PREFACE

The field of biological monitoring exists because no instrument has been devised by man that can measure toxicity! Only living material can be used effectively for this purpose. However, a biological response unaccompanied by chemical/physical data has very low information content. As a consequence, biological monitoring should be carried out in concert with chemical/physical monitoring. Although chemical/physical tests will not be emphasized in this volume, it is a *sine qua non* that all biological tests discussed are meant to be accompanied by appropriate chemical/physical tests.

For years, estimates of toxicity were based almost entirely on chemical analyses. Abundant evidence eventually accumulated that this approach was unsatisfactory. Unfortunately, biologists had not given this problem the attention it deserved and few tests were available when the need for them was recognized. However, when toxicity test methods came into favor, additional evidence quickly accumulated on the inadequacy of the use of chemical measurements alone for the determination of toxicity: (a) biological effects often occur at concentrations below analytical capabilities, (b) toxicants and other sources of stress may act quite differently in mixtures than individually, (c) environmental quality strongly mediates toxic response.

The field of biological monitoring is plagued by two needs which often appear virtually incompatible. The first is the need for replication so that experiments and tests can be repeated and validated by others and so sufficient evidence of a similar nature can be gathered for customary statistical analyses. The second is the requirement that the results be applicable and useful in the "real world", which is a highly variable complex system guaranteed to frustrate an investigator interested in replication. As one might expect, the need for replication triumphed, and the single species toxicity test carried out under highly artificial, nonvariable conditions emerged as the principal means of biological monitoring. Until relatively recently, tests were usually short term and involved lethality as an end point. In the last few years, increased attention has been given to lengthening the period of exposure and involving more than one life history stage. Even the strongest supporters of single species tests admit that the tests do not accurately reflect either the variability or the complexity of natural systems. The important question is how useful are such tests in predicting events in natural systems, particularly where pollutional effects are concerned.

Although single species toxicity tests are conducted in the laboratory, the results are generated with the intent to protect living things in natural systems. Since the assumption was made that single species tests would provide a means of estimating harm to the biota in natural systems, it is curious that the validity of this assumption has not been extensively tested in a scientifically justifiable way. There is no question that the single species test, effectively used, has reduced the number of fishkills and other catastrophic events. Nevertheless, it is regrettable that no substantive body of evidence confirming the reliability of predictions of ecosystem protection made on the basis of single species testing has been generated. Although this deficiency has been noted for years and attention continues to be called to it (e.g. Cairns *et al.*, 1981), the amount of evidence upon which one can determine the effectiveness of the single species test in predicting the response at higher levels of biological organization remains uncertain. Since prevention of harm to the environment before it occurs is the objective of ecologists, there is ample justification for developing a predictive capability that will enhance estimation of the probability of harm before material enters the environment. Determination of the accuracy of these predictions to serve as a form of error control in correcting the predictive methods is also essential. An expression of these concurrent needs for a single point discharge is depicted in Fig. 1. A variety of biological methods for carrying out both of these activities will be discussed in this book.

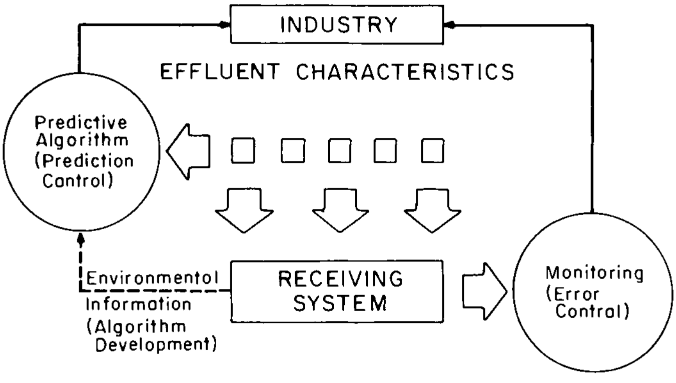

Fig. 1. Information flow in environmental control processes (from Herricks and Cairns, 1979).

Considerable unevenness exists in both the length and the level of detail in various chapters. Part I (Early Warning Systems) and Part V (Preference and Avoidance Studies) are lengthy because no comprehensive examination of the literature was available for either at the time the manuscripts were prepared. Although both fields are in early stages of development, use of early warning systems and preference and avoidance studies has been increasing rapidly in recent years and is likely to increase further in the future. Since most persons in the field of water pollution assessment may not be as familiar with this literature as they might wish, an effort has been made to cover a substantive array of publications in each of these areas. The situation is quite different in the areas covered by Part III (Community Structural Assessments) and Part IV (Toxicity Testing), since a vast amount of literature exists in both of these areas and overview articles exist. In these chapters, the strategy employed is to indicate for readers unfamiliar with these fields some earlier literature that would provide good background information. However, primary attention is devoted to important recent developments in direction and development. Both of these fields are now so extensive that even a sizeable volume would probably not adequately cover all of the important subtleties and components. As a consequence, the level of detail in methodology and so on in Parts III and IV contrasts quite sharply with Parts I and V where the areas are in earlier stages of development. Part II on functional assessments was by far the most difficult in the series to produce. This field is in an even earlier stage of development, with respect to biological monitoring at least, than any of the areas covered in this book. Although a reasonable number of candidate methods exist for determining community function for biological monitoring, none have been tested extensively for this purpose and considerable uncertainty surrounds their utility. The need for functional assessments is clear, however, and the inclusion of this chapter was primarily to affirm a conviction that this need should be fulfilled rather than an indication that an adequate information base exists. No consensus appears to exist regarding the definition of a functional assessment or what the relationship is between structural and functional attributes of an aquatic community. Despite these reservations and uncertainties, inclusion of a chapter on this subject seemed essential to call attention to the need for more extensive development of methodology. Part IV, "Future Needs," reflects my bias on the types of questions that need resolution. The chapter will serve a useful purpose if only to stimulate others to recommend alternative courses of action.

TECHNOLOGY BASED STANDARDS

Before biological monitoring is used widely both in the United States and in the world as a whole, a renewed emphasis must occur on receiving system standards as opposed to effluent standards. In the 1970s, United States federal legislation emphasized programs for removal of oxygen demanding and toxic organic chemicals from industrial and municipal wastewater discharges. The Federal Water Pollution Control Act Amendments of 1972 were designed to "restore and maintain the chemical, physical, and biological integrity of the nation's waters." This legislation required industrial discharges to meet effluent limitations by means of the application of the best practicable control technology (BPT) currently available and the best available technology (BAT) economically achievable. Pretreatment standards were also to be developed for industrial wastes discharged into publicly owned treatment works (POTW). This legislation also imposed the responsibility on the administrator of the United States Environmental Protection Agency (USEPA) to promulgate regulations restricting the discharge of toxic chemicals. USEPA did not meet all of the deadlines set and, as a consequence, was sued by several environmental groups. The settlement agreement resulting from this lawsuit required USEPA to develop a program for promulgating BAT effluent limitation guidelines, pretreatment standards, and new source performance standards for 65 chemicals and classes of chemicals. The 65 classes were subdivided into 129 specific substances now referred to as "priority" pollutants (eventually three chemicals were removed from this list).

As a result, industrial wastewater pollutants were considered in three broad categories: (1) conventional, (2) toxic (includes priority pollutants), and (3) nonconventional (those not specifically listed as toxic or conventional). BAT requirements are imposed for toxic and nonconventional pollutants, and BPT requirements are imposed for conventional pollutants. The previous discussion is intended to illustrate that wastewater effluent limitations, guidelines, and standards are primarily technology based rather than receiving system condition based. It is worth emphasizing that technology based performance standards tend to deemphasize receiving system condition and even human health. Such standards do not focus attention on and are not intended to obtain specific environmental quality. The achievement of such quality must depend on other regulations, and these must be based on the responses of the exposed organisms or surrogates for exposed organisms. Furthermore, since new properties are evident as one proceeds from single species to higher levels of biological organization (e.g. communities or ecosystems) that were not visible from an examination of the lower levels of organization, maintenance of quality depends on estimates of responses at all levels of biological organization. It is unfortunate that the 1972 Amendments to the Federal Water Pollution Control Act relegated water quality standards to a secondary role and placed primary emphasis on the technology based effluent limitations. Biological monitoring will not receive the attention it deserves until events in the receiving system are given more attention than they now are.

There are two critical deficiencies in the technology based standards: (1) the assumption that the technology utilized will always be adequate to protect human health and the environment is not scientifically justifiable, and (2) a uniform standard for the entire country fails to recognize the well established fact that the toxicity of many pollutants is a function of local water quality characteristics. This latter point has been recognized in the United States government publications (e.g. *Federal Register,* 1980, Report of the Subcommittee on Oversight and Review of the Committee on Public Works and Transportation, 1980).

VALIDATION

This book contains a variety of both field and laboratory methods. Most of the 65 criterion documents just mentioned (*Federal Register,* 1978a, b; 1979a, b; 1980) are based on single species laboratory tests with no substantive effort to validate the results in the "real world" or in complex microcosms designed to simulate important characteristics of natural systems. Validation is complicated by the failure to state more explicitly the qualities of natural systems being protected. For example, few toxicity tests involve energy transfer and nutrient cycling, yet these are very important functional characteristics of natural systems. Since some of the test species frequently used may not live in the ecosystem into which the wastes are discharged, it is obvious that extrapolations are being made. Even if a few indigenous species are used, they will almost certainly represent less than 1% of the total species being exposed. Finally, assemblages of species are known to respond differently than the same species in isolation from each other. As a consequence, it appears prudent to validate criteria and standards based primarily on laboratory tests with single species in either surrogates of complex natural systems or in the natural systems themselves. For this reason, methods at different levels of biological organization are included in this book.

ECOLOGICAL PERSPECTIVE

This book explores methods suitable for environmental management. Management is not a license to pollute, but rather the identification of desirable environmental qualities and the development of a system to maintain those qualities or achieve them if degradation has occurred. The attitude of North Americans, where vast natural areas remain relatively unchanged, toward management may not seem comprehensible to Europeans where practically all of the landscape is managed in some way. Whatever the attitudinal differences between the two groups and whatever differences now exist in the degree of management of the overall environment, the techniques discussed in this book will develop sound management practices for protecting those qualities considered particularly desirable or essential.

LAW COURTS AND SCIENCE COURTS

The legal profession in the United States has done much to call attention to environmental problems. However, the entrance in substantial numbers of the legal profession into the "environmental arena" has been a mixed blessing. In this country, at least, lawyers are advocates of the persons or positions they represent. Advocacy to this degree is not characteristic of good science which is supposed to remain objective and dispassionate in the examination of a hypothesis or a body of data. While scientists may favor a particular hypothesis, they must examine all of the evidence, pro and con, in an unbiased way (i.e. scientific objectivity) and alter their position if this seems to be justified based on evidence available. Unfortunately, many crucial environmental issues are being debated and examined in courts of law or in the framework of the law rather than the framework of science. Questions of legal precedent and the way in which evidence may be introduced dominate the examination of many environmental issues before the question or the quality of evidence and the scientific basis for the conclusion have been examined by the scientific peer review system. This review is commonplace for other scientific data outside of the environmental arena. Since money, property, lives, health, endangered species, and so on are involved and deserve the protection of the law, I do not advocate excluding the legal profession even if this were possible. However, the science on which the legal decisions are based will be more sound if provision is made for a science court to judge the science in the absence of lawyers before the evidence reaches a court of law. Because lawyers choose the form of the examination of the question, science and scientists are assigned a secondary role even where questions of scientific validity, methodology, statistics, and the like are concerned. None of the methods or strategies in this book will work well if the scientific peer review process is distorted by the legal profession. Somehow, the two professions must learn to work together on environmental problems as equals, and neither should dominate the other as is the case presently in the United States.

EFFECTIVENESS OF BIOLOGICAL MONITORING

Good evidence exists that biological monitoring may be used to document improvements in the receiving stream due to improved waste treatment methods as well as identifying the threshold beyond which further waste treatment in a particular industry does not result in substantive biological benefits (e.g. Seagle *et al.* 1980). Rickard *et al.* (1981) have described the biological colonization of an industrial pond that for 23 years had received a more or less constant flow of effluent water that at times contained radionuclides. The pond had no outlet. In this case, important societal concerns were present that could only be addressed by biological monitoring. Cushing *et al.* (1981) have used biological monitoring to show that in a river-reservoir complex the measurable body of fission-produced radionuclides decreased to essentially unmeasurable levels within 18 to 24 months of cessation of input of once-through cooling water into the river.

Although biological monitoring is not intended to replace chemical-physical monitoring, but rather should be used as an additional important line of evidence, biological monitoring may detect pollution that chemical monitoring does not. Unfortunately instances where both have been carried out for substantial periods of time are rare. Most of these are not in the open literature. However, Ruth Patrick, one of the pioneers in this field, has found such situations (personal communication).

QUALITY CONTROL

The field of biological monitoring and environmental quality control is relatively new. Thirty years ago only a few practitioners existed in a field that now has many thousands. It is inevitable that any rapidly expanding field will attract untrained people because the number of professionally competent people available is less than the demand and partly because the persons hiring environmental professionals often do not know how to evaluate background and performance. Even if this situation did not exist, quality control is mandatory; however, when a field is new and has undergone a rapid expansion, it is imperative. It is gratifying that many biological societies which once vigorously resisted certification of professions have now begun to establish procedures for doing so. While the present standards undoubtedly could be improved, at least they insure that the person certified has been exposed to certain kinds of information and perhaps even has certain demonstrated capabilities.

It is also essential that laboratories, particularly those carrying out toxicity tests, be certified on a regular basis. A one time certification is not adequate for either an individual or an organization, and recertification should occur on a regular basis.

A final form of quality control is the establishment of standard methods for gathering and analyzing data. A standard method is defined as one formally endorsed by a professional organization following established procedures. However, research should continue for improvements in these methods and for superior methods. Revision of standard methods is of course a *sine qua non*. This enhances the probability that consistency will be improved in the gathering of evidence. It will also insure that the methods used to gather data in crucial problems of environmental pollution will be methods that are better understood and more widely used than research methods. Introduction and provisional acceptance of such methods will be expedited if societies such as the Society of Environmental Toxicology and Chemistry exist where the entire membership is knowledgeable about biological monitoring and related subjects. Societies devoted primarily to other purposes may make a contribution along these lines, but it will probably be less effective because the membership as a whole is less well informed than societies primarily devoted to this subject area.

STANDARD METHODS

It is a curious fact that although a considerable amount of momentum was given to the development of toxicity testing and biological monitoring procedures by Earthday, which focused on environmental protection, the bulk of the methods used regularly for biological monitoring are either based on single species toxicity tests (which are by far the most common) or, less frequently, on diversity indices and other methods based on species counts. However, it is also worth emphasizing that only single species tests have received formal professional endorsement (i.e. as standard methods). Although I realize the difficulty of developing multispecies community and ecosystem level tests for either field or laboratory, it is unfortunate nevertheless that more such tests have not been formally endorsed as standard methods by the profession. This would insure more attention to the parameters espoused by ecologists. Merely being accepted as a standard method insures that they will be more regularly used in cases likely to go to court or even in satisfying regulatory measures. More important, businessmen are unlikely to use a method regularly until it gets the formal professional endorsement of ecologists. One can hardly blame them for this attitude–why should they invest in an experimental method not endorsed by a majority of the profession?

LITERATURE CITED

Cairns J., Jr., Alexander M., Cummins K. W., Edmondson W. T., Goldman, C. R., Harte J., Hartung R., Issensee A. R., Levins R., McCormick J. J., Peterle T. J. & Zar J. H. (1981) *Testing for Effects of Chemicals on Ecosystems.* National Academy of Sciences Press, Washington, D.C., 103 pp. Working Papers Prepared as Background for *Testing for Effects of Chemicals on Ecosystems,* 257 pp.

Cushing C. E., Watson D. G., Scott A. J. & Gurtisen, J. M. (1981) Decrease in radionuclides in Columbia River biota following closure of Hanford reactors. *Health Physics* **41,** 59-67.

Federal Register (1978a) Water quality criteria, request for comments, **43**(97), 21506-21518.

Federal Register (1978b) Water quality criteria, **43**(192), 29028.

Federal Register (1979a) Water quality criteria, request for comments, **44**(52), 15926-15981.

Federal Register (1979b) Water quality criteria, availability, **44**(144), 43660-43697.

Federal Register (1980) Proposed guidelines for deriving water quality criteria for the protection of aquatic life and its uses, **45,** 79341-79357.

Herricks E. E. & Cairns J., Jr. (1979) Monitoring and mitigation of aquatic hazard. In: *Proceedings of the Symposium on Control of Specific Toxic Pollutants* (edited by Frederick E. R.), pp. 220-231. Air Pollution Control Association, Pittsburgh, Pa.

Rickard W. H., Fitzner R. E. & Cushing C. E. (1981) Biological colonization of an industrial pond: status after two decades. *Environ. Conserv.* **8**(3), 241-248.

Seagle H. H., Jr., Hendricks A. C. & Cairns J., Jr. (1980) Does improved waste treatment have demonstrable biological benefits? *Environ. Manage.* **4**(1), 49-56.

Water Research Vol. 14. pp. 1179 to 1196
Pergamon Press Ltd 1980. Printed in Great Britain

REVIEW PAPER

BIOLOGICAL MONITORING PART I—EARLY WARNING SYSTEMS

JOHN CAIRNS JR and W. H. VAN DER SCHALIE*
Biology Department and Center for Environmental Studies, Virginia Polytechnic Institute and State University, Blacksburg, VA 24061, U.S.A.

INTRODUCTION

Civilization now faces a transitional period comparable in some ways to the one which precipitated the agricultural revolution. That revolution occurred because the unmanaged environment did not deliver food in sufficient quantity or quality to meet the expectations of human society. Mere hunting and gathering of the fruits of nature from unmanaged supplies which were subject to the vagaries of nature and, therefore, were occasionally catastrophically inadequate, were first supplemented and then replaced by managed ecosystems which came closer than nature to meeting society's expectations and needs. Similarly. we now find that the unmanaged environment is incapable of assimilating societal wastes without being seriously degraded at certain times and places. Management, not luck, is the only way to reduce such problems. Unfortunately, the frequency of unpleasant environmental perturbations and the extent of the areas affected as well as the duration of effect have increased markedly during the past few years. Not only are natural systems threatened but human health has suffered strikingly and startlingly due to mercury poisoning, kepone contamination, and a variety of other manifestations of a general problem. Moreover, the suspicion that some environmental contaminants may be influential in producing human cancer is now beginning to be supported by more substantive evidence, although this is not by any means conclusive.

Industrial societies invariably have operated on the assumption that natural ecosystems have a certain capacity for assimilating societal wastes without themselves being significantly degraded. It is all too evident that exceeding the assimilative capacity has very striking, unpleasant consequences. Unfortunately, the means of determining or estimating the assimilative capacity are not as precise as we would wish. Nevertheless, present methodology, if properly used, would certainly result in a significant and rapid improvement in our present situation, probably without the unpleasant, economic consequences that the detractors of this strategy evoke. The exciting cleanup of the Thames River and its rehabilitation as a viable fishery is too well known to most to mention here. The fact that efforts toward further cleanup have been approved recently is evidence that society feels the initial effort had a very positive cost/benefit ratio.

A crucial question for damaged ecosystems is how to determine that the improvements in effluent quality have in fact produced biological and ecological benefits. For undamaged or relatively healthy ecosystems, an important question is how to maintain quality so that no significant harm results from industrial discharges and still permit the industries to produce their products in the most efficient and least costly manner. Biological evidence is required to answer both questions for three principal reasons:

(a) Many chemical compounds and other potential pollutants produce adverse biological reactions at concentrations below present analytical capabilities.

(b) Potential toxicants are rarely present in isolation from each other. Generally toxicants are present in effluents and natural systems as a mixture, and the biological impact of the mixture cannot adequately be estimated from a series of chemical analyses alone, even if the analytical capability is adequate. In short, chemicals interact in various ways with organisms, and these interactions cannot be predicted with precision with chemical analyses alone.

(c) It is a well known fact that water quality (i.e. hardness, dissolved oxygen concentration, pH, temperature, etc.) has a very marked influence on the expression of toxicity. It is, therefore, a combination of toxicants, water quality, and the organisms present that produces a definitive estimate of the probability of harm from a specific set of concentrations and water quality conditions to a particular species. As a consequence, merely knowing the concentration of the chemical (or other potential pollutant) is not likely to produce useful management information.

The need for adequate chemical–physical data is also critical. If one only has the biological response and the water quality characteristics without knowing the concentration of the effluent or compound, the

---

This article forms one chapter of a comprehensive work on the subject under preparation by the author. This advanced publication in Water Research is by kind permission of the author, and the editor S. H. Jenkins in whom the copyright vests.

* Present address: U.S. Army Medical Bioengineering Research and Development Laboratory, Fort Detrick, MD 21701, U.S.A.

correlation between concentration and response cannot be determined. Therefore, adequate information on dose-response curves must include an array of information about: (a) the species of organisms tested; (b) the water quality and other test conditions; and (c) the concentrations of the chemicals or other potential pollutants being tested (Cairns *et al.*, 1978).

It is well established that both water and effluent quality are not constant. Water quality may fluctuate daily, or even hourly, and certainly fluctuates widely seasonally as well. Regional differences in water quality are so well established that no further documentation is needed. It is established also equally well that effluent quality and quantity also vary, but do so according to production schedules that are societally controlled rather than under the influence of natural forces. It is obvious, therefore, that the receiving capacity of natural systems will not cycle in phase with the fluctuations in effluent quality or quantity. A prime management need is a means of determining ecosystem assimilative capacity for societal wastes on a site specific basis. The biological means of doing this in a systematic way constitutes the developing field of biological monitoring. Although chemical–physical monitoring will not be discussed in detail in this series of papers, it is a *sine qua non* that this type of monitoring must accompany the biological monitoring and be correlated with it. The need for this type of quality control system with a coupling of biological and chemical-physical sensors both "in plant" and "instream" (e.g. the receiving system) have been discussed in detail elsewhere (Cairns, 1975a,b; Cairns *et al.*, 1972, 1973a,b). The essence of these environmental quality control systems is the use of biological parameters to estimate the health of the organisms in the receiving system or anticipate damage to these organisms by a variety of predictive methods (i.e. early warning systems). Tentatively the methods just mentioned will be as follows:

Part I—Biological Monitoring—Early Warning Systems.

Part II—Biological Monitoring—Receiving System Methodology Based on Biological Function.

Part III—Biological Monitoring—Receiving System Methodology Based on Community Structure.

Part IV—Biological Monitoring/Toxicity Testing.

Part V—Biological Monitoring—Preference and Avoidance Studies.

Part VI—Biological Monitoring—Overview of Future Research Needs and Directions.

The essence of the entire field of biological monitoring is that one cannot protect the health, condition, or quality of a natural system without obtaining information directly about the condition of that system and the organisms that inhabit it. Furthermore, the organisms must not only be able to survive but be able to function normally as well. As a consequence, one needs an array of information based on diverse and dissimilar methodologies in order to have a reasonable expectation of adequately protecting the ecosystem receiving the potential pollutant. If this information is not gathered on a systematic basis, it would not fulfil the requirements of a quality control system. The field of biological monitoring was developed in order to control and maintain effectively environmental quality at socially and biologically desirable levels.

Over the past 20 or 30 yr those concerned with environmental quality have searched for a single all purpose method of measuring environmental health or condition. This is the contemporary version of the search for the holy grail and almost certainly will be no more successful. Nevertheless, one constantly sees papers with criticisms that a particular method (such as a diversity index) does not provide all the information necessary about the condition of a biological or ecological system. *No one method ever will!* The realization of this simple fact, although far from universal, has resulted in the production of a series of protocols which are merely a systematic way of gathering the information necessary to make a sound decision on the hazard to human health and the environment as a consequence of using a particular chemical or discharging a certain type of waste. A representative source list for this information will shortly be published (Dickson *et al.*, 1979), and some earlier versions are already available (Cairns & Dickson, 1978; AIBS, 1978).

One continually encounters the question: Why should I bother with biological monitoring since it was never necessary in the past? It is a simple fact that water is no longer an economical "free good." The following quotation illustrates this point.

> With ever increasing demands being placed upon limited water resources, it has become evident that in most of the United States water has become a scarce resource; scarce in the sense that one use will affect other uses. It must now be recognized that competition for water is a fact, that tradeoffs must be considered seriously, that in some cases there must be restrictions on use (and therefore development), and that water is no longer the "free good" that once was taken for granted. (U.S. Water Resources Council, 1978).

As a consequence of the removal of quality water from the "free goods" category, its use now has a price tag. One of the components of this price tag is biological monitoring now required in United States of America by various enacted legislation. It should be evident to industry and other water users that the funds allocated to biological monitoring are not totally lost. They will provide an economic benefit because the information generated will tell when the assimilative capacity is being underutilized as well as when it is in danger of being overutilized (full discussion of the assimilative capacity concept is in Cairns, 1977). Since the assimilative capacity is not constant, a systematic way of tracking its changes involving biological monitoring is an essential component of water quality control in an enlightened industrial society.

One aspect of biological monitoring is the use of aquatic organisms to provide an early warning of the presence of toxic materials in water. Possible applications of this concept in an industrial situation are to help prevent hazardous waste spills or in a water treatment plant as a check on potable water supplies. These tasks traditionally have been carried out exclusively by chemical–physical techniques applied either continuously or at frequent intervals. The inadequacy of these methods by themselves in predicting toxicity has already been indicated. This article describes the operational requirements which must be met by a biological toxicity early-warning system and some of the organisms and techniques which have been or may be employed in such systems. An early warning toxicity monitoring system will be considered to have the following characteristics:

1. The organisms are held either in a laboratory situation or in the field under controlled conditions and are exposed on a frequent or continuous flow basis to the water or wastewater being tested.
2. A physiological or behavioral parameter of the organism is monitored by a recording device with the capability of responding to abnormal conditions indicated by the organism.
3. The function of the monitor is primarily for detection of short-term changes in toxicity as opposed to chronic or cumulative effects of a toxicant.

DISCUSSION

The idea of using aquatic organisms for continuous toxicity monitoring is not new. One early type of monitoring system used fish placed in flowing water or wastewater (Henderson & Pickering, 1963; Jackson & Brungs, 1966). The fish were to be observed visually for mortality or signs of stress. In another system used in Sweden since 1965, fish were exposed to diluted waste from cellulose plant, and their condition was observed several times daily. This approach has helped in determining the source of toxic effects (Hasselrot, 1975).

Visual monitoring of lethal effects has the obvious drawback of requiring that someone be present continually to observe the organisms. Moreover, there may be a considerable delay between the onset of toxicity and death. Consequently, the current emphasis in early warning systems is on automated devices which measure some prelethal symptom of poisoning, such as abnormal respiration or activity. This may allow toxicant-induced responses to be detected sooner and with greater sensitivity.

While the number of potential early warning systems is large, each one must meet certain conditions if it is to be useful. These constraints need to be considered whether one is reviewing a current monitoring system or designing a new one. The following list of requirements includes suggestions given by Poels (1975, 1977), Ladd (1977), and Brown (1976).

1. The physiological or behavioral parameter of the organism selected for monitoring should be quantifiable through appropriate interfacing techniques for analysis either by a computer or other electronic recording equipment. This will enable the operation of the system to be both continuous and automatic. However, the method itself should not result in undue stress on the organism. Techniques requiring restraint of the organism or the attachment of devices to it may be less desirable for this reason.
2. Rapid, reliable detection of developing toxic waste conditions is of prime importance. The speed with which an organism will react is influenced by a large number of variables. These include the type of organism and the particular response being monitored, the concentration of the material with respect to acutely toxic levels, the toxicant's mode of action, and the physical–chemical characteristics of the dilution water (temperature, pH, dissolved oxygen, etc.). Table 1 gives some response times for parameters which either have been or could be used in monitoring systems. Delays of several hours between introduction of a toxicant and a reaction by organism being tested may not be rapid enough to allow prevention of a toxic waste spill unless there is a built-in delay between exposure of the organism and the escape of the toxicant (Cairns *et al.*, 1972; Price, 1978). Long-term effects caused by low levels of materials with cumulative toxicity (for example arsenic or some pesticides) are not likely to be detected soon enough for the response to be useful (Brown, 1976).

The reliability of the monitoring method chosen should be such that the system will respond repeatedly to the presence of a variety of toxic materials. While it may be possible to select an organism that is sensitive to several toxicants in a particular industrial waste effluent, it is unlikely that any single organism could respond at the proper level to the range of chemicals in drinking water that might be harmful to man (Brown, 1976). Price (1978) cited data that indicated wide differences between European potable water quality criteria and the sensitivity of one current toxicity monitor which measures fish ventilatory rates (Morgan, 1977).

Loss of sensitivity to toxicants may occur following long-term exposure to very low levels of the toxic material. Bluegill sunfish (*Lepomis macrochirus*) exposed for 29 weeks to zinc at 1/100 of the 96-h LC50 (0.075 mg l^{-1}) showed some decrease in activity responses to a simulated zinc spill (3.0 mg l^{-1} zinc). On the other hand, ventilatory responses were not reduced even after a 41-week pre-exposure. Acclimation following a response to sublethal toxicant levels may occur also. Increases in the coughing rate of brook trout (Drummond & Carlson, 1977) and the oxygen consumption of bluegill sunfish (O'Hara, 1971a) peaked and began to return to pre-exposure levels within 24 h after the start of exposure to sublethal concentrations of copper. These types of prob-

Table 1. Response times of some aquatic organisms to various toxicants

Response time	Toxicant level	Response criteria	Organism	Reference
Several minutes	55,115 $\mu g\,l^{-1}$ copper (96 h LC50 = 115 $\mu g\,l^{-1}$)	Increase in coughing frequency	Brook trout (*Salvelinus fontinalis*)	Drummond *et al.* (1973)
Approx 15 min	pH 3.4, pH 11, 10 $mg\,l^{-1}$ copper, 10 $mg\,l^{-1}$ cyanide	Increase of 1 $mg\,l^{-1}$ in effluent dissolved oxygen level	Biological filtration unit (microbial)	Solyom *et al.* (1976)
25 min (death at 2 h)	150 $\mu g\,l^{-1}$ Lindane	Loss of rheotaxis	Rainbow trout (*Salmo gairdneri*)	Poels (1977)
45 min (death at 4–8 h)	60 $\mu g\,l^{-1}$ Lindane	Loss of rheotaxis	Rainbow trout	Poels (1977)
<1 h	0.5, 2.5 $mg\,l^{-1}$ cadmium	Abnormal activity levels in two of four crayfish	Crayfish (*Cambarus acuminatus*)	Maciorowski *et al.* (1977)
<1 h	2930 $mg\,l^{-1}$ acetone (24-h LC50 = 6100 $mg\,l^{-1}$)	Increased ventilation rate and buccal pressure amplitude	Rainbow trout	Majewski *et al.* (1977)
1 h	Peak of 6200–6800 $mg\,l^{-1}$ acetone (96-h LC50 = 8300 $mg\,l^{-1}$)	Abnormal ventilatory rates in three of four fish	Bluegill sunfish (*Lepomis macrochirus*)	van der Schalie *et al.* (1979)
1 h	0.1 $mg\,l^{-1}$ cyanide	Reduction of 36% in efficiency of nitrification	Biological nitrification column (microbial)	Stroud & Jones (1975)
<2 h	15 $\mu g\,l^{-1}$ copper	Elevation in serum cortisol levels	Coho salmon (*Oncorhynchus kisutch*)	Schreck & Lorz (1978)
2–4 h	≤48-h LC50 of: cadmium, copper, magnesium, lead, mercury, phenol, ammonia, cyanide, carbamate, chlordane, parathion, pentachlorophenol	High ventilatory rates from 60% or more of the fish tested	*Micropterus salmoides*	Morgan (1977)
3–10 h	0.8 of the 96-h LC50, bleached Kraft mill effluent	Elevation in plasma glucose levels	Coho salmon, Rainbow trout	McLeay (1977)
11 h	4.16 $mg\,l^{-1}$ zinc	Elevation in ventilatory rate	Bluegill sunfish	Cairns & Sparks (1971)
<24 h	6.0 $\mu g\,l^{-1}$ endrin	Elevation in coughing rate	Bluegill sunfish	Drummond & Carlson (1977)
41–44 h	0.1 $mg\,l^{-1}$ cadmium	Abnormal activity levels in two of four crayfish	Crayfish (*Cambarus acuminatus*)	Maciorowski *et al.* (1977)
52 h	2.55 $mg\,l^{-1}$ zinc	Elevation in ventilatory rate	Bluegill sunfish	Cairns & Sparks (1971)
>240 h	0.4 $\mu g\,l^{-1}$ endrin	Elevation in coughing rate	Bluegill sunfish	Drummond & Cairns (1977)
1/4 of the LT50	DDT	Maximal time to loss of swimming ability in 50% of test fish	Carp	Besch *et al.* (1977)
1/2 of the LT50	Mercury	Maximal time to loss of swimming ability in 50% of test fish	Carp	Besch *et al.* (1977)

lems may be minimized by replacement of the organisms monitored at regular intervals.

3. A monitoring system should have a minimum of false alarms-responses to nonharmful variations in water quality. Certain characteristics of water (or wastewater) such as temperature, pH, dissolved oxygen, or hardness may cause responses from organisms when no specific toxicant is present, or they may make a given amount of toxic material more or less harmful. (Even responses to some toxicants may not be desirable; residual chlorine present in many drinking water supplies would have to be removed before the water could be used in a toxicity monitoring unit.) Bluegills in a pollution monitoring system showed increased breathing and activity rates when the diurnal temperature cycle was changed from a range of 24.8–26.0°C to a range of 24.8–29.2°C. A similar system did not respond to a nontoxic change in calcium levels from 10 to 107 $\mathrm{mg\,l^{-1}}$ (Cairns *et al.*, 1973a,b, 1974). Opercular, coughing, and metabolic rates of young rainbow trout (*Salmo gairdneri*) were all affected by sublethal variations in pH between pH 6 and pH 9 (Hargis, 1976). Fluctuations in dissolved oxygen levels are likely to have a direct effect on the operation of pollution monitoring systems measuring oxygen consumption or operacular rates. Some regulation of these water quality paramcters, or at least knowledge of their values, are necessary if proper conclusions are to be drawn about the cause of an abnormal reaction by an organism.

4. Appropriate methods for the analysis of data must be developed. The normal range of variation in the parameter being monitored should be statistically determined so that reliable criteria can be established for abnormal responses caused by toxic conditions. When individual organisms are monitored, variations between individuals make it advisable to use several organisms and to have each serve as its own control. (A separate set of control organisms also may be appropriate.) Control data obtained after acclimation to test conditions could be used to generate confidence intervals by which abnormal responses could subsequently be detected. This approach has been used in monitoring systems which utilize fish activity patterns (Cairns *et al.*, 1973a, b; Hall *et al.*, 1975) and breathing patterns (Cairns *et al.*, 1973a, b; Morgan & Kuhn, 1974). When the parameters being monitored have a diurnal periodicity, it may be necessary to compute separately a normal range of values for several different periods of the day.

5. Monitoring systems should be relatively easy to operate and should produce results which are easy to interpret. This would not be difficult for an electronic system in which all data analysis and most control functions could be done automatically. A relatively simple electronic device could, for example, turn on an alarm light when it determined that toxic waste conditions were developing. Highly trained personnel would not be needed to run such a system.

6. The organism used in the monitoring system should be fairly inexpensive and easy to acquire. This limits the selection of species considerably since relatively few are commercially available. Advantages of using standard test organisms include the availability of toxicity literature and culture techniques and not having to continually modify monitoring systems for each new species. On the other hand, it may be desirable, when monitoring waste effluents, to use an organism common to the body of water receiving the waste.

7. The monitoring apparatus should be reliable and require as little maintenance as possible. Environmental control (temperature, humidity, etc.) may be necessary, especially if electronic components are involved. Complex mechanical arrangements should be kept to a minimum. It should be possible to develop biological monitoring systems that are comparable in cost to physical–chemical monitoring systems.

The basic design of an early warning biological monitoring unit might include a water or wastewater delivery system, experimental chambers, electronic or mechanical data transducers feeding into a data analysis system, and an alarm system to provide notice of developing toxic conditions. The type of transducer used will depend on the biological parameter being monitored. This device could be an amplifier which magnifies the microvolt signal generated by fish as they ventilate their gills or the electrical output of an oxygen electrode which measures oxygen consumption. Interfacing most electrical signals to a small computer could be done easily using standard techniques. Commercial multichannel data acquisition systems are available for this purpose.

The choice of a suitable organism and physiological or behavioral parameter for monitoring is most important. In describing some of the many possibilities below, emphasis will be on techniques using fish, although a number of methods have been developed for invertebrates. Willingham & Anderson (1966, 1967) suggest several possible means of using microorganisms to detect toxic materials in water. The possibility of continuously monitoring the phototactic response of microcrustaceans such as *Daphnia* and *Artemia* is discussed by Willingham and Anderson, as continuous-flow bacterial systems based on measurement of bioluminescence or oxygen uptake.

A complex automated water monitoring system, now undergoing testing by the United States National Aeronautics and Space Administration, utilizes three bacterial biomass monitoring devices along with conventional physical-chemical sensors (Jeffers & Taylor, 1977, Taylor & Jeffers, 1977). Total bacteria counts (living and dead) are determined by measuring light from a chemiluminescent reaction catalyzed by the porphyrins from lysed bacterial cells. An estimate of living bacterial biomass is found by assaying ATP (adenosine triphosphate) levels from lysed cells. This is done by measuring light emitted in a bioluminescent process powered by the ATP. (The reaction uses